



# **ANNUAL REPORT**

**July 1, 2019 to June 30, 2020**

# Table of Contents

---

<b>Preface</b> .....	Page 3
<b>Introduction to the Annual Report</b> .....	Page 5
<b>Institutional Information</b> .....	Page 12
- Research Summaries	
- Key Personnel	
<b>Project Progress Reports</b> .....	Page 47
- Project Progress Reports by Institution	
<b>2019-2020 Publications, Manuscripts, &amp; Grants</b>	
- Publications and Manuscripts.....	Page 226
- Current and Pending Grants .....	Page 267
<b>Scientific Abstracts</b> .....	Page 316
<b>Institutional Budgets and Justifications</b> .....	See companion report

## Preface

The COVID-19 pandemic is upon us, and it has had a profound impact on us all. It has already taken major tolls on our safety, well-being, economic stability, work and way of life. It has impelled us to find new ways to protect and address the needs of our patients, research participants and families, colleagues, communities and each other, avert an overwhelming healthcare program, and prepare for the challenges and uncertainties that lie ahead. It has led us to rearrange our clinical and research priorities and rise to the immediate challenges while finding ways to address our longstanding goals. It has called on us to do so from a social distance, yet do it together.

Like others around the world, researchers, clinicians and organizations in the Arizona Alzheimer's Consortium continue to respond to this rapidly responding crisis in ways that put everyone's safety first, adhere to the social distancing precautions needed to dampen the virus's spread, and reduce the chance that scarce healthcare resources will be overwhelmed. At the same time, we have sought to fulfill the stated objectives of the Consortium and its National Institute on Aging (NIA) supported Arizona AD Center (ADC) as much as possible. We have modified or temporarily suspended many studies, sought to communicate virtually with our patients, participants and families, and reserved in-person visits for those activities that are deemed essential to our stakeholder's safety and well-being.

Due to the social distancing precautions, we canceled the Annual Scientific Conference, which the University of Arizona was scheduled to host in May, we plan to conduct our annual External Advisory Committee Meeting virtually in June, and we sought and kindly received permission from the Arizona Department of Health Services (ADHS) to complete several projects after the standard June 30 deadline. Despite these precautions, we continue to make great progress, and we have included this year's project progress reports and research abstracts, publications, and grants in this Annual Report, submitted our annual non-competing ADC progress report to NIA, begun to prepare for our five-year competing ADC grant application in September, and plan to hold a smaller conference for our young investigators when appropriate later this year. While it was necessary to cancel this year's Annual Conference, it was a painful decision. We are pleased to report that the University of Arizona has again agreed to host next year's conference and that Dr. Zaven Khachaturian (the galvanizing force behind development of the infrastructure for AD research and a valued advisor to our consortium) has again agreed to provide the Leon Thal Memorial Lecture at that time. Finally, we will evaluate the possible need to reschedule our annual Public Conference, which normally takes place in October, if indeed circumstances warrant a restructuring or rescheduling of this important event.

Like everyone else, I have been astounded by the myriad of ways in which you, our clinical and research colleagues, have worked together to prioritize the safety and well-being of our patients, research participants, and their families, including our particularly vulnerable older adults, as well as the ways you have been there for each other. I have been moved by your compassion, creativity, sacrifice and "in this together" spirit, and by the courage of our clinical colleagues on the front lines. You have responded to this crisis with uncommonly common heroism, you have embraced the opportunity do so, and you have found ways to further address our longstanding goals along the way.

I am extremely grateful to my clinical and research colleagues, patients, research participants and families in the Arizona Alzheimer's Consortium for all you do to make us proud. You have found ways to address the needs of our patients and research participants, while continuing to make major contributions to the study of Alzheimer's disease (AD), related disorders and the aging brain, and you continue to give us a chance to achieve our ambitious goals.

My Consortium colleagues could not be more grateful to our Arizona Department of Health Service leaders, elected officials, and organizational leaders for their enthusiastic and steadfast support. You have shown extraordinary support in financially good times and bad, including during the current crisis. You have recognized the need to make a difference in the fight against AD, shared our sense of urgency, and given us the chance to do so right here in Arizona.

While not contagious, AD and related dementias have been described by the World Health Organization and other policy leaders as the coming 21<sup>st</sup> century pandemic, a ticking time-bomb that is projected to affect well over 100 million families around the world within the next 20 years due to the growing number of people living to older ages. Together, we have a chance to help transform the understanding, treatment and prevention of AD, and we have a fighting chance to do so before 2025.

No matter how disruptive or long-lasting, we will do our part to respond to the COVID-19 crisis, we will continue to play leadership roles in the effort to avert the coming AD pandemic, and we will do so right here in Arizona.

With best wishes for everyone's health, safety and well-being during this challenging time,

A handwritten signature in black ink that reads "Eric M. Reiman, M.D." The signature is written in a cursive style with a large, stylized 'E' and 'R'.

Eric M. Reiman, M.D.  
Director, Arizona Alzheimer's Consortium



## Introduction to the Annual Report

### Background

The Arizona Alzheimer's Consortium is the nation's leading model of statewide collaboration in Alzheimer's disease (AD) research. It includes more than 150 researchers and staff from seven principal organizations, including Arizona State University, Banner Alzheimer's Institute, Banner Sun Health Research Institute, Barrow Neurological Institute, Mayo Clinic Arizona, the Translational Genomics Research Institute, and the University of Arizona, and from four affiliated organizations, including the Critical Path Institute, Midwestern University, Northern Arizona University and the University of Arizona College of Medicine, Phoenix. Established in 1998, the Consortium is intended to make a transformational difference in the scientific fight against AD, to engage Arizona's underserved and understudied Native American and Latino communities, to help address the unmet needs of patients and family caregivers, and to advance the understanding and promotion of healthy cognitive aging. The Consortium's major themes are the early detection and prevention of AD. Its primary goal is to find effective AD prevention therapies as soon as possible.

The Consortium is widely recognized as a model of multi-institutional collaboration in biomedical research. It capitalizes on complementary resources and expertise from different disciplines and organizations to address scientific problems in the most impactful way. Its researchers receive critical support from the state of Arizona (through the Arizona Department of Health Services [ADHS]), the participating organizations, a competitive Arizona AD Core Center (ADCC) grant from the National Institute on Aging (NIA), and numerous other grants and contracts.

Eric M. Reiman, MD, is the Director of the Consortium and the NIA-sponsored ADCC, Richard Caselli, MD, is the ADCC's Associate Director, and Carol Barnes, PhD, chairs the Consortium's 26-member Internal Scientific Advisory Committee. Mr. David Jerman is Administrative Director of the Consortium's state- and organizational-supported research program, Mrs. Andrea Schmitt is Administrative Director of its ADCC grant, and Executives from each of the seven principal organizations serve on the Consortium's Board of Directors. The Consortium's external advisors include Drs. Marilyn Albert, Zaven Khachaturian, Bruce Miller, and Thomas Montine, who are internationally recognized for their contributions and leadership roles in the study of AD and/or related disorders. They conduct annual site visits, review the progress and productivity of the Consortium and ADCC, and provide formal feedback and recommendations to the researchers, NIA, and state.

The Arizona Alzheimer's Consortium capitalizes on the state's strengths in brain imaging and emerging fluid biomarkers, genomics, the computational, mathematical and statistical analysis of complex data sets, the basic, cognitive and behavioral neurosciences, clinical and experimental therapeutics, and neuropathology research. It has made pioneering contributions to the scientific understanding, unusually early detection and tracking of AD, the accelerated evaluation of putative AD prevention therapies, and the scientific understanding of the aging mind and brain. It has introduced new ways for different stakeholders to work together, it has provided data, biological samples and interested research participants for researchers inside the state and

around the world, and it has introduced promising new care models for patients and family caregivers. It continues to attract new researchers and clinicians, and support other biomedical research developments in the state. Indeed, it has helped to make Arizona a destination center for the advancement of AD research and care.

State and organizational matching funds continue to provide the “glue” for this geographically distributed research program, the “fuel” needed to launch new research initiatives, and the framework needed to reach the Consortium’s over-arching goals. Funds are used to support dozens of research projects each year, almost all of which involve researchers from different scientific disciplines, and about half of which include researchers from different organizations. As one of our advisors observed, Arizona has become known around the world for its courage, groundbreaking organizational and scientific paradigms, and ability to make things happen in the fight against AD.

The Arizona ADCC has received continuous competitive NIA grant funding since 2001. The ADCC’s Administrative, Clinical, Data Management and Statistics, Neuropathology, and Outreach and Recruitment Cores, a Research Education Component (REC), and a competitive Pilot Project Program have supported researchers and projects inside and outside of the state. In July 2016, the ADCC received its fourth consecutive five-year renewal grant, after being noted for its exceptional track record, productivity and impact, its outstanding scientific contributions, regional, national, and international initiatives and impact, its effective leadership and collaborative model, impressive commitments from the state and each of our participating organizations, and its leadership roles in the fight against AD. In July 2018, the ADCC received a competitive supplement to establish a critically needed Brain Imaging and Fluid Biomarker Core.

### **Productivity, Progress and Impact**

The Arizona Alzheimer’s Consortium is the leading statewide AD Center in the nation and one of the most productive AD research programs in the world. Since its inception in 1998, its researchers have generated thousands of publications, grants and contracts, and more than \$2 billion in new investments. Consortium researchers have made pioneering contributions to the study of AD, related disorders, and the aging mind and brain:

1. They have helped to clarify genetic and non-genetic (e.g., microbial) risk, resilience and resistance factors and disease mechanisms, offered targets at which to aim new AD treatments, provided new insights about the pathological changes associated with AD and related disorders, and introduced promising new ways to treat and prevent AD.
2. During the past year, they implicated a rare APOE variant in resistance to autosomal dominant AD, characterized the biomarker effects and potential mechanisms that might account for this resistance, demonstrated that APOE and its variants have a greater impact on the differential risk of AD than previously thought, and introduced new opportunities for the study of APOE and development of APOE-modifying gene and drug therapies.
3. They continue to generate invaluable public resources of longitudinal, neuropathological, and gene expression data for the field; they have begun to provide a new resource of DNA and RNA sequencing data from different brain cell types and regions in brain donors with and without AD; and they have used these and other resources to implicate disease networks, risk factors, and potential drivers at which to aim new AD treatments.
4. They continue to introduce new data-sharing, biological sample-sharing and collaborative paradigms to assist researchers in Arizona and around the world—including data and samples from their own observational studies and prevention trials, data from a growing number of clinical trials of AD and other disorders through the Critical Path for AD (e.g., CPAD, [6](https://c-</a></li></ol></div><div data-bbox=)

[path.org/programs/cpad/](http://path.org/programs/cpad/)); and on-line memory tests and other information that has been generated in >110,000 participants in a program called MindCrowd ([www.mindcrowd.org](http://www.mindcrowd.org)).

5. They and their colleagues have played leadership roles in the early detection and tracking of AD, including the detection and tracking of progressive brain imaging, other biomarkers, and cognitive changes—as well as the detection of neurodevelopmental changes—in cognitively unimpaired persons at genetic risk, and they have provided invaluable resources of data and volunteers from persons at three levels of genetic risk for AD (i.e., with two, one and no APOE4 alleles) and in Colombian autosomal dominant AD (ADAD)-causing mutation carriers and non-carriers from the world's largest ADAD kindred. They introduced new experimental paradigms, image-analysis techniques and composite cognitive tests to help in this endeavor. Their work anticipated and advanced the conceptualization of preclinical AD. As noted below, this work continues to inform the design of prevention trials in persons at increased genetic and/or biomarker risk and provides the foundation needed to launch a new era in AD prevention research.

6. They continue to clarify how different molecular processes and brain cells, regions, networks, and mental operations orchestrate memory and other thinking abilities, and how they are affected by AD and aging. They have developed, tested and used groundbreaking neuroscientific, experimental and behavioral paradigms to help in these endeavors; and they have played leading roles in the international study of the aging mind and brain.

7. They have played leadership roles in brain imaging and other research studies to detect, track and study AD and related disorders starting many years before the onset of symptoms, assess genetic and non-genetic risk factors, and introduce image analysis methods to address these goals with improved power. They have played leadership roles in the effort to validate amyloid and emerging PET methods in persons at the end of life who subsequently donate their brains and support future FDA approval for their use in the clinical setting. They have begun to develop resources and tools to put promising cerebrospinal fluid (CSF) assays, blood tests, and mobile technologies to the test as soon as possible.

8. They and their collaborators have recently used PET to demonstrate an absence of amyloid plaque burden and presence of tau tangle abnormalities suggestive of chronic traumatic encephalopathy (CTE) and the absence of AD in living former professional football players, and have major roles in “Diagnose CTE”, the national study in former professional and college football players.

9. They continue to provide a world-leading scientific resource of longitudinal and neuropathological data, brain and body tissues for the study of AD, Parkinson's disease, and related disorders in their Brain and Body Donation Program—and they have begun to incorporate ante-mortem biomarkers and new brain tissue resources to help researchers address their goals with even greater impact.

10. During the past year, they used this resource to support the use of plasma p-tau measurements in the differential diagnosis of AD, helping to set the stage for the use of this and other blood-based biomarkers in research, drug development and clinical settings.

11. With support from NIA, philanthropy and industry, their Alzheimer's Prevention Initiative (API) continues to play leadership roles in the accelerated evaluation of promising prevention therapies. API's public-private partnerships include the following international trials: 1) The API ADAD Colombia trial of the anti-amyloid antibody therapy crenezumab in the world's largest ADAD kindred; 2) the API Generation Study 1 of a BACE inhibitor (an oral anti-amyloid production drug) and active immunotherapy in cognitively unimpaired APOE4 homozygotes; 3) the API Generation Study 2 of the same BACE inhibitor in additional cognitively unimpaired APOE4 homozygotes

and amyloid-positive APOE4 heterozygotes; 4) a planned API/A4 AD Prevention Trial in cognitively unimpaired amyloid positive adults; and 5) future trials in the early planning stages. The API ADAD Colombia Trial is on track to be completed in 2022. Medications in Generation Studies 1 and 2 were recently discontinued due to early modest and non-progressive cognitive worsening (an apparent class effect); the trial will end in the next few months to confirm that those effects are reversible; and the trial will provide a public resource of data and samples in an unprecedented number of APOE4 homozygotes and heterozygotes. The API/A4 trial will provide a fighting chance to find and support the accelerated approval of a plaque-reducing prevention therapy by 2025 and introduce a more rapid way to evaluate prevention therapies using surrogate biomarker endpoints.

12. API also includes exceptionally large registries and related programs to support enrollment in AD prevention trials and related studies. It includes a Colombian API Registry with nearly 6,000 mutation carriers and non-carriers from the world's largest ADAD kindred, the North American Alzheimer's Prevention Registry with ~350,000 members ([www.endALZnow.org](http://www.endALZnow.org)), GeneMatch (a national resource of more than 85,000 members who permitted us to characterize their APOE genotypes for research purposes), genetic risk disclosure and impact assessment programs to help support interest and enrollment in prevention trials; engagement programs to inform participants about relevant prevention trials and other research opportunities; and other emerging methods and strategies to help find and support the approval of an AD prevention therapy as soon as possible. These and related efforts have had a profound impact on researchers, policy makers, and other stakeholders around the world.

Consortium researchers continue to develop groundbreaking research methods and strategies, collaborative models and data, and biological sample-sharing paradigms to support these and other research endeavors. They continue to capitalize on their ADCC Cores, other shared resources and collaborations to assist in this effort. Furthermore, they continue to conduct state-supported collaborative research studies to advance new ideas, find those that have the greatest impact, and generate the findings, publications and grants to have the maximum public impact. They continue to generate new findings, publications in the highest impact medical and scientific journals, and competitive grants and contracts for the study of AD, related disorders and brain aging. They continue to make historic contributions to AD research, and they have generated the resources and collaborations needed to recruit and support a growing number of researchers and trainees to our participating institutions.

During the past year, consortium researchers have generated groundbreaking findings, published them in some of the world's leading scientific and medical journals (e.g., the Nature journals and the New England Journal of Medicine), and have had a major impact on the field. Their contributions to the understanding of APOE and its variants on the development and potential treatment and prevention of AD, the roles of emerging blood-based biomarkers in the diagnosis, prognosis, early detection, tracking, treatment and prevention of AD, the potential role of microbial pathogens in the development of AD, and recent CTE findings promise to have a major impact on the field. Our researchers have continued to demonstrate the value of applying big-data analyses to the genome-wide analysis of inherited and expressed genes in brain donors with and without AD and advancing push-pull relationships between big-data analyses of omics data from human research participants and experimental data from cellular and animal models to discovery of AD mechanisms, risk factors, and new treatments. They continue to secure major grants, contracts, and philanthropic investments (including two major gifts totaling \$60M in the last year alone), recruit new researchers at most of our organizations, develop new clinical programs, and seek to address the needs of our under-represented and under-served Latino and Native American communities. They continue to advance current and new AD prevention trials, provide better tests of the amyloid hypothesis, anticipate the development and testing of APOE-

modifying treatments, and provide the best chance to find an effective prevention therapy by 2025.

### **Challenges, Opportunities, and New Initiatives**

Despite global progress in AD research, the past year has been marked by disappointing findings from several trials of anti-amyloid treatments, primarily in clinically affected patients. Nearly 100% of AD clinical trials have failed since 2002, causing stakeholders to ask what comes next. Here, we briefly summarize some of the challenges, opportunities and ongoing or planned initiatives we have in mind to address them.

A fighting chance to find and support the approval of a prevention therapy by 2025. As previously noted, our researchers continue to provide better tests of the amyloid hypothesis in cognitively unimpaired persons at increased genetic or biomarker risk, including persons at genetic risk who do not yet have significant amyloid plaque deposition. They continue to incorporate novel experimental paradigms in their trials to have the greatest impact, and they have a chance to find and support the approval of a prevention therapy by 2025 and support the possible qualification of biomarker endpoints in 24-month prevention trials.

Diversifying the portfolio of promising AD treatments. Our researchers are taking a multi-faceted approach to address this urgent need: 1) As briefly noted in last year's report, we are using high-quality brain tissue from our Brain and Body Donation Program to develop a public resource of RNA and DNA sequencing data from different brain cell types and regions in 100 AD cases and controls, and we are actively involved in the development and analysis of complementary omics data in the Accelerated Medicines Partnership for AD (AMP-AD). We have recruited leaders in the big-data analysis of these omics data sets, such that we can discover multi-scale networks, drivers of these networks, and repurposed or new drugs for this fundamentally human disease, and put them to the test in relevant cellular, animal, and other experimental models. Conversely, we can use the human data sets to clarify the extent to which novel findings from the experimental models are relevant to the human disease. This push-pull relationship between experimental and human data, along with close collaborations with other groups, will be a defining feature of two of our developing translational research programs. 2) We and our collaborators have capitalized on the study of an ADAD mutation carrier who was resistant to AD dementia to further inform the role of APOE and its variants in the predisposition, resistance, and resilience to AD, support the potential value of APOE silencing gene therapies in the treatment and prevention of AD, suggest novel approaches to the development of APOE-related gene and drug therapies, and set the stage to rapidly put APOE modifying treatments to the test when they become available within the next 2-3 years. We have extended this approach to the study of APOE4 homozygotes and heterozygotes who are resistant to AD dementia. 3) We have also begun to capitalize on extremely large electronic health record (EHR) data sets to support the identification of repurposed drugs and disease mechanisms. 4) Our researchers continue to explore other approaches to the treatment and prevention of AD in translational, and to a lesser extent, early phase studies including those that target metabolic, mitochondrial, tau, neuroinflammatory, and/or hormonal processes. We have commitments to support the recruitment of researchers at several Arizona institutions. 5) There will eventually be opportunities to extend API's prevention trials to emerging anti-tau therapies and combination therapies.

Appealing to our industry partners to remain engaged in the fight against AD and related diseases. Now more than ever, public-private partnerships and NIA funding are needed to give makers of promising treatments the courage and conviction to stay the course despite so many disappointments in clinical trials and do so in innovative yet rigorous ways. We are well positioned to advance that effort in both the drug discovery and drug development effort.

Brain Aging Research. Arizona researchers continue to play a leadership role in the study of the normal aging brain and the continued promotion of healthy aging. This effort is reflected by

the UA's McKnight Research Program, a wide range of studies in unimpaired older and younger adults, non-human primates, laboratory rodents, and other models, studies of aging in the MindCrowd Program, promising drug development efforts, and a recently submitted U19 grant to support further advancement of these and other ambitious efforts.

Dramatically increasing the value of our cohorts. While we follow several important research cohorts in our longitudinal studies and prevention trials, the value of our cohorts would be dramatically increased by the incorporation of biomarkers and biological fluid samples to characterize amyloid, tau, neurodegenerative and cerebrovascular disease burden, and, when available, other neurodegenerative (e.g., synuclein and TDP-43) pathologies. Unfortunately, brain imaging biomarkers are too expensive to use on all of our participants, lumbar punctures for existing and emerging CSF assays are relatively invasive and somewhat limited in availability, and promising blood tests remain to be further clarified. We have invested in several initiatives to address this challenge. We have recently played major roles in the evaluation of emerging blood-based biomarkers, and have begun to develop strategies to establish their roles in research, drug development and clinical settings.

Increasing the study of our under-represented Native American and Hispanic research participants. We continue to explore ways in which to increase participation of these research participants in our ADCC Clinical Core and other research programs. 1) We have begun to capitalize on interactions with the Strong Heart Stroke Study and University of Washington AD Center Native American Satellite Core, contribute to the acquisition of genetic and MRI data, analysis of brain imaging, other biomarker and cognitive data, and mentorship of young investigators. 2) We are working with (and play a leadership role in) the UA-Banner All of Us Research Program, which has already enrolled >26,000 persons, including >8,400 and 1,400 Hispanic and Native American participants, respectively. 3) We plan to develop and maintain an active cohort of at least 100 Native American and 100 Hispanic research participants in our clinical core by July 2021.

Promoting the development of new investigators. Consistent with the new round of Requests for AD Research Center (ADRC) grant applications, we have placed a growing emphasis on, recruitment and mentorship of new investigators, including young student and faculty investigators, established investigators who are new to our field, and a growing number of investigators from under-represented groups. Our programs include our NIH-supported post-doctoral and pre-doctoral research training programs, support for competitive pilot study grant applications, courses in the conduct of relevant research studies, other outreach, educational and research internship programs for students from a wide range of ages and backgrounds, and support for their participation in relevant conferences (whenever they can resume) and our annual retreat. We will further develop these research education and training programs over the next year.

New programs, researchers and physicians. We are pleased to announce the opening of BAI Tucson, extending its comprehensive care model and working in partnership with UA to advance AD, Lewy Body Dementia (LBD) and aging research in well characterized research participants. We have been privileged to receive major donations to advance AD research and care at several of our institutions. We continued to increase the number of physicians and productive researchers at several of our institutions.

Coping with COVID. As noted in the Preface, the COVID-19 crisis has had a major impact on our clinical and research programs, the way in which we interact with our patients and research participants, and just about everything else in our lives. Like other researchers, clinicians, and organizations around the world, we are working hard to respond to the crisis, support the ability of our health systems to cope with COVID-19, and continue to address other critical goals. We

recognize that we are only at the beginning of what could be a long and historically challenging time. But we are determined to find ways to adapt and learn from the current crisis (e.g., in terms of how to capitalize on virtual research and clinical communications in more effective ways) and stay on target as we continue to advance the fight against AD.

### **Looking Ahead**

During the next few years, we and our colleagues will continue to develop several new scientific and clinical initiatives. We will continue to lead and expand our AD prevention trial programs. We will continue to develop public resources of data and biological samples to advance the study of AD, and support the development of promising CSF and blood-based biomarkers of AD and related disorders. We and our colleagues continue to advance the study of CTE in former football players and develop a shared resource of electronic health record data and biological samples from 100,000 persons in the All of Us Research Program, including a large proportion of persons from Hispanic, Native American and other under-represented groups. We will continue to find new ways to advance the understanding and promotion of healthy brain aging, develop new models of dementia care, provide a foundation to discover a more diversified portfolio of promising treatments, and provide the best fighting chance to find and support the approval of a prevention therapy by 2025. We will begin to advance the use of blood-based biomarkers in each of these endeavors. We are extremely grateful to the state of Arizona, NIA, our participating organizations, colleagues, collaborators, advisors, research participants, and other supporters. We are proud of our progress and excited about our plans. Together, we are determined to make a transformational difference in the fight against AD—and do so despite the challenges we all face during the COVID-19 pandemic.

## **Institutional Information**

### **Research Summaries and Key Personnel From Each Participating Institution**

# ARIZONA STATE UNIVERISTY

## Institutional Abstract

Over a decade ago, ASU set forth to redefine higher education by focusing on a model of the New American University. With swift momentum, ASU has led the world with innovative ideas to student-centric public higher education, honing in on academic excellence, the highest quality education and training, inclusiveness to a broad demographic, and maximum societal impact. Underscoring this exemplary new path, ASU has been ranked number one for innovation by U.S. News and World Report for the last five years (2015-2019). With Alzheimer's disease affecting roughly one in nine people 65 years old and over, and one in three people 85 years old and over, research on Alzheimer's disease exemplifies the type of endeavor that ASU seeks to promote, and a focus on innovative approaches is most certainly critical to research and treatment efforts.

For the Arizona Alzheimer's Consortium, ASU provides the Outreach and Recruitment Core and Research Education Component. These serve researchers throughout the state as part of the Consortium's NIA-sponsored Arizona Alzheimer's Disease Center. The ASU team includes leaders in the development of novel models to: establish a causative link between traumatic brain injury (TBI), neuroinflammation, and Alzheimer's disease (Brafman laboratory); explore injury-induced neuroinflammation as a contributor to Alzheimer's Disease (Stabenfeldt laboratory); characterize the microbiome of post-mortem brain tissue in subjects affected by Alzheimer's disease (Readhead laboratory); address the overlapping and unique expression profiles among neurodegenerative diseases (Mastroeni laboratory); evaluate sex differences and gonadal hormone contributions to the trajectory of behavioral and neuropathological change across aging (Bimonte-Nelson laboratory); investigate sex differences in healthy and Alzheimer's Disease brain gene expression (Wilson laboratory); study age-related changes in neurobehavioral underpinnings of nicotine addiction vulnerability in females across age groups (Gipson-Reichardt laboratory); conduct computational image analysis and implement biomathematical techniques to increase the power to detect and track Alzheimer's disease progression (Wang laboratory); and, develop and test multicomponent interventions for individuals living alone with MCI (Coon research laboratory). It is noteworthy that ASU has numerous scientific research domains that are being further developed and strengthened to bolster the impact on Alzheimer's disease and aging research, with a focus on discovery and action to move trajectories, diagnosis, and treatment forward. These include, but are not limited to, the neurosciences, health outcomes research, and focused translational research realms that pose hypothesis-driven questions approached from a systems and interdisciplinary perspective. Collectively, ASU has a solid framework and wide-ranging strengths that are poised to make great strides in the scientific fight against Alzheimer's disease, as well as to optimize the trajectory of brain aging, using both preclinical and clinical approaches. Moreover, it is noteworthy that the assets in the research programs at ASU within the Arizona Alzheimer's Consortium represent a range of colleges, institutes, and centers across ASU.

ASU and Phoenix-based Banner Health, one of the nation's largest nonprofit health systems, have launched a research alliance to advance the scientific study, treatment and prevention of Alzheimer's, Parkinson's and other neurodegenerative diseases. The partnership includes the establishment of the Neurodegenerative Disease Research Center<sup>1</sup>. The center is an extension of the partners' work with the Arizona Alzheimer's Consortium and is envisioned to become one

---

<sup>1</sup> <https://science.asu.edu/neurodegenerative-disease-research-center>

of the world's largest basic science centers for the study of Alzheimer's and other neurodegenerative diseases. The Center is expected to grow to include about 20 new laboratories and additional affiliated laboratories. It will foster push-pull relationships between big data and other analyses of post-mortem and other human data sets and experimental models and leverage an emerging collaboration among several consortium partners to provide a public resource of detailed omics data from different cell types and regions in clinically and neuropathologically characterized brain donors. The Center is intended to further clarify disease mechanisms and risk factors for AD and related disorders, provide new therapeutic targets, and support the discovery of new treatments and biomarkers.

A strength of ASU is the training, mentoring, and education of future generations of aging and neurodegenerative disease researchers and academicians, spanning high school students, to undergraduate students, to graduate students, to postdoctoral fellows. The approach to training is hands-on, multifaceted, and interdisciplinary, with the goal to engage future scientists in aging and neurodegenerative research to yield maximal impacts on research discovery and translational outcomes. The new Research Education Component, Co-Directed by Dr. Heather Bimonte-Nelson (ASU), reflects this strong and extensive training commitment. Notably, ASU offers graduate degrees in Statistics and Biomedical Informatics, the Behavioral Neuroscience Program<sup>2</sup> within the Department of Psychology, as well as the Interdisciplinary Graduate Program in Neuroscience<sup>3</sup>. The latter two training programs focus upon approaches that integrate multiple levels of analysis using systems and interdisciplinary approaches – cellular, behavioral, and cognitive – to address preclinical, clinical, and translational questions about brain and behavior relationships.

---

<sup>2</sup> <https://psychology.clas.asu.edu/content/psychology-behavioral-neuroscience-phd>

<sup>3</sup> <https://neuroscience.asu.edu>

# ARIZONA STATE UNIVERISTY

## Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on Project</b>
Ahmed, Kinza	--	Undergraduate Research Assistant
Andrew, Kieran	--	High School Student
Angulo, Aylin	BS	Research Specialist
Arroyo, Ana	--	Undergraduate Research Assistant
Bandin, Eric	--	Undergraduate Research Assistant
Bimonte-Nelson, Heather	PhD	PI; Professor
Brafman, David	PhD	PI; Assistant Professor
Brookhouser, Nicholas	MS	Graduate Researcher
Bulen, Haidyn	--	Undergraduate Research Assistant
Bull, Amanda	--	Lab Technician
Carbajal, Berta	--	Research Specialist/Promotora
Carll, Phil	MSW	Research Coordinator
Christian, Jordan	--	Undergraduate Research Assistant
Coon, David W.	PhD	PI; Professor
Copeland, Connor	BS	Graduate Research Assistant
Cordova, Lourdes	--	Survey Interviewer/Promotora
Cortes, Marielysse	MSW	Program Manager
Cutts, Joshua	MS	Graduate Researcher
Delzepich, Sascha	--	Undergraduate Research Assistant
Evanovich, Austin	--	Research Assistant
Fan, Yonghui	MS	Graduate Researcher
Farazi, Mohammad	MS	Graduate Researcher
Gipson-Reichardt, Cassandra	PhD	Principal Investigator; Assistant Professor
Glinka, Allison	MS	Research Specialist
Goldman, Jami	MSW	Research Coordinator
Gomez Morales, Abi	MS	PhD Student
Johnson, Raena	--	Undergraduate Research Assistant
Knittel, Jacob	BS	Research Technician
Koebele, Stephanie	PhD	Post-doctoral Researcher
Kostes, William	BS	Graduate Researcher
Leyrer-Jackson, Jonna M.	PhD	Postdoctoral Fellow
Logan-Robledo, Santiago	--	Undergraduate Research Assistant
Manzo, Alyssa	--	Undergraduate Research Assistant
Mason, Christopher	PhD	Co-investigator
Mastroeni, Diego	PhD	PI
Mroczek, Megan	--	Undergraduate Research Assistant

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on Project</b>
Munoz, Ivan	--	Undergraduate Student
Nguyen, Toan	BS	Graduate Researcher
Northup-Smith, Steven	--	Lab Manager
Oddo, Salvatore	PhD	Associate Professor
Opachich, Zorana	--	Undergraduate Research Assistant
Overby, Paula	--	Lab Manager
Peay, Dylan	--	Conrad Lab Graduate Student
Pena, Veronica	--	Graduate Student
Peters, Mollie	MS	Research Assistant
Piña, Jose	--	Undergraduate Research Assistant
Prakash, Shefali	--	High School Student
Raman, Sreedevi	MS	Graduate Researcher
Ramsey, Jaden	--	Undergraduate Research Assistant
Readhead, Ben	MBBS	Principal Investigator
Srinivasan , Gayathri	MS	Graduate Researcher
Stabenfeldt, Sarah	PhD	PI, Associate Professor
Stotler, Kassey	MEd	Research Specialist
Strouse, Isabel	--	Undergraduate Research Assistant
Ta, Duyan	MS	Graduate Researcher
Tekel, Stefan	BS	Graduate Researcher
Tu, Yanshuai	MS	Graduate Researcher
Van Do, Ngoc	--	Undergraduate Research Assistant
Vijayaraghavan, Shalini	--	Undergraduate Research Assistant
Wang, Yalin	PhD	Associate Professor
Willingham, Crystal	BS	Laboratory Manager
Wilson, Melissa	PhD	Principal Investigator
Witten, Amanda	MS	Research Technician
Woner, Victoria	--	Graduate Student
Wu, Jianfeng	BS	Graduate Researcher

# **BANNER ALZHEIMER'S INSTITUTE**

## **Institutional Abstract**

The Banner Alzheimer's Institute (BAI) has three goals: To find treatments to prevent Alzheimer's disease (AD) without losing a generation, to set a new standard of care for patients and families, and to promote a model of multi-institutional collaboration in biomedical research. BAI is intended to accelerate the evaluation, approval and availability of treatments to postpone, reduce or completely prevent the clinical onset of AD as quickly as possible; leverage its brain imaging resources and expertise to advance the scientific study, early detection, tracking, diagnosis, treatment and prevention of AD and related disorders; address the medical and nonmedical needs of affected persons and families to the fullest extent possible, and help to establish a new standard of dementia care in the emerging population-based healthcare financing system. Finally, it is intended to complement, enhance, and benefit from close working relationships with its organizational partners inside and outside of the Arizona Alzheimer's Consortium (AAC).

BAI's Stead Family Memory Center includes a Memory Clinic, Family and Community Services Program and Clinical Trials Program. It offers a wide range of services for the evaluation and care of affected persons and family caregivers, helping to address their medical and non-medical needs throughout the illness. It provides educational, outreach and research enrollment programs for Arizona's Native American and Latino communities, evaluates and follows Native Americans in the NIA-sponsored Arizona AD Center's Clinical Core, and oversees an Annual Conference on AD and Dementia in Native Americans. Its Banner Dementia Care Initiative is seeking to demonstrate ways in which to optimize the identification and evaluation of cognitive problems, address a broad range of the affected person's and family's medical and non-medical needs, reduce unnecessary hospitalizations, and is affordable to payers in the emerging healthcare financing system. BAI conducts numerous clinical trials of investigational treatments, including those in the Alzheimer's Prevention Initiative (API). Its researchers also help oversee an NIA-sponsored cohort study of cognitively unimpaired persons with two, one and no copies of the APOE4 allele, which has helped to conceptualize the preclinical stages of AD, an NINDS-sponsored study of chronic traumatic encephalopathy (CTE) in former National Football League and college football players, and one of the Precision Medicine Initiative's (PMI's) first healthcare provider-led cohort programs in a partnership between University of Arizona and Banner Health.

Its state-of-the-art NIH-supported Imaging Center includes two PET/CT systems, a 3T MRI, cyclotron, radiochemistry laboratory, and computational image analysis laboratory. It provides imaging resources and expertise, research PET tracers, image-analysis methods, data and biological samples for researchers inside and outside of Arizona. In collaboration with Mayo Clinic Arizona, it includes a longitudinal brain imaging study of cognitively unimpaired persons with two copies, one copy, and no copies of the APOE4 allele, reflecting three levels of genetic risk for late-onset AD, and image-analysis techniques with improved power to characterize subtle brain changes over time. In collaboration with the University of Antioquia and a Harvard post-doctoral student, it also includes a study of PSEN1 E280A mutation carriers and non-carriers from the world's largest autosomal dominant AD kindred in Colombia. It is a member of the AD Neuroimaging Initiative (ADNI) PET Core, where it is responsible for the development, testing and use of voxel-based image analysis techniques with improved power to detect and track AD. It has played pioneering roles in the study of preclinical AD. AARC funds complement research activities supported by competitive grant awards from several NIA-sponsored research grants, private foundation grants, and clinical trials. In conjunction with our NIA-sponsored ADCC, subjects, images, other data, and image-analysis techniques from our study of cognitively normal

APOE ε4 carriers and non-carriers provide a core resource for interested investigators inside and outside of Arizona.

In early May 2020, BAI announced the opening of BAI Tucson. The 10,000 sq. foot memory center will offer comprehensive services for patients and families and conduct research studies into the treatment and prevention of memory disorders under the leadership of Dr. Allan Anderson and in collaboration with colleagues from the University of Arizona.

In the next few years, BAI, BSHRI, and their partners will place a growing emphasis on the acquisition of antemortem brain-imaging, CSF, and blood-based biomarkers for AD and related disorders in their longitudinal cohorts, and help to find and support the use of promising amyloid and other blood tests for AD and related disorders. These organizations, TGen, and ASU (e.g., at the ASU-Banner Neurodegenerative Disease Research Center [NDRC]) are also developing a shared resource of DNA and RNA sequencing data from different brain cell types and regions in high-quality brain samples from AD cases and controls and are using big data analytical techniques to characterize networks and drivers at which to target in the discovery of new treatments. They and their organizational partners will also be exploring targets at which to aim APOE modifying treatments.

With several hundred million dollars in NIH, philanthropic and industry support, API has helped to launch a new era in AD prevention research, accelerate the evaluation of prevention therapies, and help to find and support the approval, availability and affordability of prevention therapies as soon as possible. It includes a growing number of preclinical AD / theragnostic biomarker development trials in persons who, based on their genetic or biomarker findings, are at increased AD risk, including the API ADAD Colombia Study in the world's largest autosomal dominant AD (ADAD) kindred, the international API Generation Studies 1 and 2 in persons at particularly high risk for the clinical onset of late-onset AD, a NIH supported API-A4 Alzheimer's prevention trial in cognitively unimpaired amyloid-β positive adults and other trials TBD. The trials are intended to evaluate the investigational treatments in potentially license-enabling prevention trials; to provide a better test of the amyloid hypothesis than trials in the later preclinical or clinical stages of AD; establish the extent to which a treatment's different biomarker effects are associated with a clinical benefit and provide evidence to support their use as potential surrogate endpoints in future 24-month prevention trials; provide a shared resource of data and biological fluids for the research community after the trial is over; complement, support and provide a foundation for other prevention trials; help clarify the benefits, risks and role of APOE genetic test disclosure in the era of Alzheimer's prevention trials; support the advancement of Alzheimer's prevention research in the Collaboration for Alzheimer's Prevention (CAP); empower persons at highest risk in the scientific fight against AD; and provide a fighting chance to find and support approval of an AD prevention therapy by 2025.

API also includes exceptionally large registries to support interest and possible enrollment in prevention studies. In partnership with the University of Antioquia, the API Colombian Registry now includes ~6,000 members of the PSEN1 E280A mutation kindred, including nearly 1,200 mutation carriers, who have provided their DNA and have had clinical and neuropsychological evaluations. The web-based Alzheimer's Prevention Registry ([www.endALZnow.org](http://www.endALZnow.org)) now provides information about advances in prevention research and opportunities to enroll in prevention trials to >350,000 people and continues to grow rapidly; our GeneMatch Program ([www.endALZnow.org/genematch](http://www.endALZnow.org/genematch)) has enrolled >85,000 persons and aims to enroll >100,000 persons 55-75 years of age, match interested participants in API and other prevention trials and begin to clarify what it means to learn about one's APOE test results; and these programs continue to grow. It continues to champion new ways to identify and support enrollment in prevention trials

(e.g., using an amyloid- $\beta$  blood tests), and to address the logistical, ethical, and scientific issues involved in this endeavor.

BAI has several specific aims:

1. To leverage our imaging resources in the early detection, tracking, and diagnosis of AD, the clarification of genetic and non-genetic risk factors, and other collaborative research studies inside and outside of Arizona.
2. To leverage our imaging resources in the early detection and tracking of related diseases (e.g., chronic traumatic encephalopathy [CTE]).
3. To implement, test and use PET radiotracer techniques (e.g., for the assessment of amyloid and tau pathology) in the study of AD and related disorders.
4. To develop image analysis techniques and composite cognitive test scores with improved power to detect and track AD and evaluate AD-modifying and prevention therapies.
5. To accelerate the evaluation of AD prevention therapies through API's preclinical AD trials and enrollment registries.
6. To share data and biological fluid samples with the research community, establish a public resource of blood samples from thousands of well characterized persons, help the field develop and evaluate blood tests for AD and related disorders as soon as possible, and advance the complementary research goals of our partners inside and outside Arizona.
7. To provide a care model that fully addresses the needs of patients and families and BAI, and to develop and test the cost-effectiveness of a dementia care program that better addresses the needs of patients and family caregivers in the Banner Health Accountable Care Organization in the Banner Dementia Care Initiative.
8. To support the clinical research and Native American outreach, education and enrollment goals of the Arizona ADCC.
9. To promote the further development, productivity, and close working relationships of research programs involved in the fight against AD and related disorders.

# BANNER ALZHEIMER'S INSTITUTE

## Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on project</b>
Reiman, Eric	MD	Executive Director, BAI Director, Arizona Alzheimer's Consortium (AAC) and NIA-supported Arizona ADCC
Tariot, Pierre	MD	Director, BAI
Amador, Ricardo	MS	ADCC Data Coordinator
Bandy, Dan	MS, CNMT	PET Technical Director and Sr. Scientist
Boker, Connie	BS, MBA	Director, Imaging Center Operations
Burke, William	MD	Former Director, Stead Family Memory Center
Chen, Kewei	PhD	Sr. Scientist, Computational Image Analysis
Copeland, Jacquelynn	PhD	Neuropsychologist
Craig-Muller, Jennifer	BS	Clinical Research Program Senior Manager
DeMarco Kathryn	BS	Clinical Research Program Manager
Ghisays, Valentina	PhD	Post-Doctoral Fellow; Bioinformatics Scientist
Goldfarb, Danielle	MD	Physician Neurologist, Memory Center
Gopalakrishna, Ganesh	MD	Associate Director, Stead Family Memory Center
Goradia, Dhruvan	PhD	Bioinformatics Scientist
High, Nellie	MS	Research Project Coordinator
Jaeger, Chad	BS	Research Administrator Senior Director
Jakimovich, Laura	RN	Multi-Center Clinical Trials Manager
James, Michelle	PsyD	Neuropsychologist
Jansen, Willemijn	PhD	Post-Doctoral Fellow (Part-Time)
Koren, Andrei	PhD	Senior Scientist, Lab Head Radiochemistry Research
Langbaum, Jessica	PhD	Associate Director, Alzheimer's Prevention Initiative
Langlois, Carolyn	MA	Clinical Research Program Manager
Lee, Wendy	MS	Assistant Director, Computational Brain Imaging
Lomay, Nicole	BS	Native American Outreach Representative
Patel, Roma	MS	Clinical Trials Senior Manager
Malek-Ahmadi, Michael	PhD	Bioinformatics Scientist
Nisson, Lori	MSW/ LCSW	Director, Family & Community Services
Pandya, Sachin	BS	Clinical Research Coordinator
Perrin, Allison	MD	Physician Dementia Specialist
Protas, Hillary	PhD	Bioinformatics Scientist
Saner, Don	MS	Senior Director, Data Science; Director, ADCC Data Management and Statistics Program
Su, Yi	PhD	Director, Computational Brain Imaging Analysis Program; Co-Director, ADCC Data Management and Statistics Program
Tsai, Po-Heng	MD	Physician Dementia Specialist, Memory Center
Weidman, David	MD	Director, Stead Family Memory Center

# BANNER SUN HEALTH RESEARCH INSTITUTE

## Institutional Abstract

**Banner Sun Health Research Institute (BSHRI)** was established in 1986 in the heart of Sun City, Arizona, the nation's first planned retirement community, including more than 100,000 older adult residents in the area, and intended to make a profound difference in the scientific study of Alzheimer's disease (AD) and Related Dementias (ADRD), Parkinson's disease (PD), other age-related brain disorders, and healthy aging.

BSHRI includes: **a)** A world-renowned **Brain and Body Donation Program (BBDP)** for the study of AD/ADRD, PD, related disorders, cancer and aging; **b)** **Comprehensive, multidisciplinary and integrated clinical centers and programs** in cognitive, memory and movement disorders that provide coordinated world-class care and services that include subspecialist clinicians and staff from The Cleo Roberts Memory and Movement Centers, The Division of Neuropsychology, Family and Community Services, and the Neurowellness Program; **c)** More than 30 ongoing NIH, foundation, and biopharma-sponsored state-of-the-art **clinical trials and observational cohort studies** for AD/ADRD, PD and movement disorders and cognitive aging; **d)** The **Center for Healthy Aging**, with a Longevity Longitudinal Cohort Study of nearly 1,489 research participants (641 active), including 242 individuals of age 85 or older and 124 individuals of age 90 years or older, for the study of cognitive aging; as well as a free, community service Brain Health Check-In (BHCI) Program (>430 BCHIs performed since established in December 2018) to provide walk-in or scheduled brain health concern assessments along with feedback, information, education, resources and referrals; **e)** Extensive **outreach, education, training and volunteer programs** including >130 education programs per year (nationally, internationally, regionally and locally) and leadership in world-renowned continuing education programs; training in neuropsychology for students and post-doctoral fellows; neurology residents; a highly productive summer research internship program for under-represented and other college and high school students, and partnerships with Sun Health Foundation and other stakeholders in this highly concentrated community of active older adults; **f)** Leadership roles and close working collaborations and relationships with AD/ADRD and movement disorders consortia, clinicians, scientists, educators, public health advocacy groups and organizations throughout Arizona and around the world. Where historically, the state's largest number of productive basic scientists in the fight against AD, who are well-known for their major contributions to the study of amyloid and tau processing, brain inflammation, epigenetics, and the roles of cholesterol and cerebrovascular disease in AD, were located; these basic science programs have now completed relocation to ASU. From July 2001 to June 2016, BSHRI served as the applicant organization for the Arizona ADCC on behalf of the organizations in the Arizona Alzheimer's Consortium, and it remains home to the ADCC's Administrative Director, Andrea Schmitt.

The world renowned BBDP, directed by Thomas Beach, MD, PhD, includes ~ 900 actively followed, clinically characterized and longitudinally assessed participants, including patients with AD, PD, and related disorders, and older adults with cancer who are cognitively and neurologically unimpaired at the time of their enrollment. All participants consent to donate their brains and/or bodies after death. The BBDP is unique for: **a)** its rapid autopsy program, with a median 3-hour post-mortem interval allowing unusually high tissue quality, optimizing post-mortem discovery research on the >2,000 expired donors, who have had comprehensive neurological assessments during life and neuropathological examinations after death; **b)** the unusually large number of brain donors who are cognitively and neurologically unimpaired at the time of their clinical enrollment, thereby advancing the study of preclinical AD and PD and providing numerous clinically and

neuropathologically normal control subjects for genetic and other research studies; **c)** whole body donation, banked organs and tissues from >700 expired donors since 2005, and the opportunity to relate brain pathology to biological features of other body organs; and **d)** approximately 200 annual tissue distributions to advance research in Arizona and around the world. The BBDP includes many research participants in the Arizona ADCC's Clinical, Neuropathology and Brain Imaging and Fluid Biomarker Cores, in partnership with Mayo Clinic Arizona and Barrow Neurological Institute. In addition, it continues to play critical roles in the neuropathological validation of amyloid PET, tau PET, and other ante-mortem biomarker measurements in end-of-life (e.g., hospice) patients, thus contributing to FDA approval of molecular imaging/PET measurements in the clinical setting. The BBDP continues to provide a tissue resource for genome-wide genetic, transcriptomic and proteomic data from different brain regions and cell types, and to contribute to numerous research studies, collaborations, grants, and dozens of annual publications and impactful findings.

Since 2016, BSHRI has undergone significant changes, shifting focus from basic sciences to clinical and translational science and clinical services, and setting the stage for BSHRI and its organizational partners to further develop its AD/ADRD, PD and movement disorders, and aging clinical, research, education, training and outreach programs. These changes include: **a)** Ongoing harmonization of Banner Alzheimer's Institute's AD/ADRD-related clinical, family and community services, clinical research and clinical trials programs on its downtown Phoenix and BSHRI campuses including launch of the Dementia Care Partners community care navigation and support program; **b)** Further growth of comprehensive and integrated multidisciplinary services at The Cleo Roberts Memory and Movement Disorders Centers including recruitment of several clinicians/clinician-scientists; **c)** Successful implementation (with AAC pilot funding to PI Dr. Danielle Goldfarb, previously at Banner Alzheimer's Institute, now full-time at BSHRI; Dr. Alireza Atri, Co-I) of an ultrasound lumbar puncture (LP) program (58 LPs performed as part of the program since July 2019); **d)** Successful launch and expansion of the Brain Health Check-In (BHCI) community service program at the Center for Health Aging; since December 2018 these walk-in or scheduled BHCI have provided > 430 individuals with free brain health concern status assessments along with feedback, information, education, resources and referrals; (Featured in 2019 on Channel 10 Fox News; see link: <http://www.fox10phoenix.com/news/arizona-news/free-test-at-banner-can-let-you-see-how-healthy-your-brain-is>); **e)** Substantially enhancing clinical and biological (biofluid/serum) characterization of the BSHRI's Longevity Study cohort (see current AAC report of pilot funding in FY 2019-20, Dr. Alireza Atri PI), and harmonizing important elements and increasing co-enrollment in the Longevity Study and BBDP programs; **f)** Ongoing strategic planning for the development and further growth of clinical, aging and clinical/translational research programs, services, and training and education programs on the BSHRI campus -- in addition to BSHRI's large clinical, family and community services, PD-related "NeuroWellness", and clinical trials programs, its scientific, education and outreach efforts include >130 international, national, regional, and community presentations per year; BSHRI staff provided >12,000 person/hours of medical/health professional education, scientific or community lectures, presentations and programs, including co-sponsoring and co-directing (Dr. Atri) the world-renowned Harvard Medical School annual 4-day CE course (Dementia: A Comprehensive Update <https://cmeregistration.hms.harvard.edu/events/dementia-a-comprehensive-update/event-summary-ff22ba91677f4bc8b0df67daffb28240.aspx?dvce=1>); and **g)** Expanding the BBDP in impactful ways, including achieving an increase of enrollment to ~900 annually assessed prospective brain donors; inclusion of blood, CSF and/or imaging data and samples in many BBDP participants; and development of a public resources of sorted cells and omics data from different cell types and regions that differ in the vulnerability and resilience to elements of AD pathology enabling us and our TGen, NDRC, consortium colleagues, and other researchers to better clarify disease networks, and new treatment targets.

# BANNER SUN HEALTH RESEARCH INSTITUTE

## Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on project</b>
Adams, Debra	--	Research Scheduler, Center for Healthy Aging
Arce, Richard	--	BBDP Pathology Technician
Atri, Alireza	MD, PhD	Director, Banner Sun Health Research Institute
Auman, Briana	PysD	Neuropsychologist
Beach, Thomas	MD, PhD	BBDP & Neuropathology Core Director, Neuropathologist
Beh, Suet Theng	PhD	Postdoctoral Fellow
Belden, Christine	PsyD	Neuropsychologist
Burks, Teresa	NP	Nurse Practitioner
Bunkley, Latasha	--	Clinical Research Assistant
Callan, Michael	--	Clinical Research Assistant
Cipriani, Dana	--	Clinical Research Rep
Cline, Carol	--	Psychometrist Coord
Davis, Kathryn	BA	Clinical Core Coordinator, ADCC and BBDP
De Santiago, Stephanie	DNP	Nurse Practitioner
Dhanani, Sara	MD	Movement Disorders Neurologist
Glass, Michael	--	BBDP Pathology Technician
Goldfarb, Danielle	MD	Neuropsychiatrist (dual Neurologist/Psychiatrist)
Intorcica, Anthony	--	BBDP Pathology Technician
Keane, Marissa	--	Clinical Research Assistant
Kiefer, Jamie	--	Psychometrist
Liebsack, Carolyn	RN, BSN	Clinical Trials Program Operations Director Center for Health Aging
Long, Kathy	--	Clinical Research Rep
Lue, Lih-Fen	PhD	Senior Scientist, Human Cells Core for Translational Research, BBDP
MinerRose, Daneva	--	Clinical Research Coordinator
Moorley, Naudia	PsyD	Neuropsychologist
Nelson, Courtney	--	BBDP Pathology Technician
O'Connor, Kathy	MS	Outreach Program Manager/Longevity Program Coordinator
Oliver, Javon	--	BBDP Pathology Technician
Papa, Jaclyn	--	BBDP Pathology Technician
Powell, Jessica	PsyD	Neuropsychologist
Quinones, Patricia	--	Research Scheduler
Rangel, Amy	--	Phlebotomist
Reade, Marina	FNP	Nurse Practitioner
Rich, Maggie	--	Psychometrist
Roye, Lisa	--	Psychometrist

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on project</b>
Russell, Aryck	--	BBDP Pathology Technician
Sahagun, Anela	--	Research Scheduler
Schmitt, Andrea	BS, CRA	ADCC Administrative Director
Schofield, Sharon	--	Research Data Specialist
Serrano, Geidy	PhD	Anatomist Supervisor, BBDP
Shaikh, Farah	--	Clinical Research Assistant
Shprecher, David	DO	Movement Disorders Program Director; Neurologist
Spann, Bryan	DO, PhD	Dementia Neurologist
Sue, Lucia	BS	Coordinator and Tissue Donation Manager, BBDP
Teran, Marlene	--	Clinical Research Assistant
Vargas, Daisy	--	BBDP Pathology Technician
Walker, Jessica	--	BBDP Pathology Technician
Zamrini, Edward	MD	Former Director, Memory Clinic

# **BARROW NEUROLOGICAL INSTITUTE at St. Joseph's Hospital and Medical Center Institutional Abstract**

The Barrow Neurological Institute focuses on human and animal research that can translate to clinical care. The BNI focus in Alzheimer's Disease and aging is in prevention, early detection and defining mechanisms of AD. Investigators at Barrow Neurological Institute engage in human subject studies including clinical trials and laboratory science research of human nervous system function in health and disease processes that can translate into improvements in clinical care. Barrow's work related to Alzheimer's disease and aging concerns new treatment intervention to combat human cognitive decline, early detection of dementing disorders, and identification or refutation of hypothesized cellular and molecular mechanisms in AD.

These studies also cross over to additional work on other neurodegenerative disorders. In the past few years, neurodegenerative disease research at Barrow has expanded with the addition of both accomplished senior faculty members and more junior investigators with promise and skill and new ideas about disease mechanisms and treatment opportunities. Laboratory and clinical resources devoted to this enterprise also have increased, and further growth in this area is planned and expected.

The close relationships between clinicians and scientists mean that many cross-disciplinary studies are underway or being developed at Barrow. The Alzheimer's Disease and Memory Disorders Program has seen a significant increase in patient clinic visits in the past year, which enhances recruitment of patients for research. AAC support has allowed for the development of the **Hispanic Enrollment in Alzheimer's Research Trials** (the **HEART** Program) at BNI which is focused on engaging underserved and understudied populations in clinical research, as well as establishment of the necessary infrastructure to engage, retain, and recruit Latinos.

Funding increases, generously matched and exceeded by Barrow resources, allowed for expansion of pilot research project awards, including new lines of research in advanced multiparametric magnetic resonance imaging techniques in mild cognitive impairment and aging, molecular and cellular mechanisms related to nucleocytoplasmic trafficking deficits of ADAR2 and RNA editing aberrations in Alzheimer's disease, roles  $\alpha 7$  nicotinic acetylcholine receptors ( $\alpha 7$ -nAChRs) in mediated chronic A $\beta$  exposure-induced neurotoxicity, neural hyper-synchronization and disease neuro-pathogenesis, and neural changes associated with Down's syndrome. Funding has also allowed for the development of a biobank focused on collecting biofluids and development of cell lines from Alzheimer's and Frontotemporal dementia patients, providing an extremely valuable resource to the research community.

BNI scientists include accomplished senior faculty members with extensive expertise in neurodegenerative disorders, who promote cross-departmental and cross-institutional collaborations in their investigation of treatment opportunities.

Dr. Rita Sattler's laboratory focuses on the elucidation of neurodegenerative disease mechanisms in ALS, FTD and other Alzheimer's disease related dementias. Based on a strong translational research background, including time spent working for a small biotech

startup company, Dr. Sattler uses patient-derived induced pluripotent stem cell (iPSC) models of varying subtypes of neurons and glia cells to study cellular and molecular changes that occur during disease manifestation and progression with the ultimate goal of identifying novel therapeutic targets. Recent studies are focused on the role of RNA binding proteins ADAR2 and TDP-43 and their contribution to aberrant disease-mediated RNA editing and splicing, respectively. In addition, the Sattler laboratory has established iPSC microglia-neuron co-culture models to study the impact of the neuro-immune axis on cortical neuronal degeneration observed in dementias, including FTD and AD. Finally, Dr. Sattler is greatly interested in the mechanisms of synapse damage and loss which is observed in AD and FTD, but also other neurodegenerative disease accompanied by cognitive impairments, such as ALS and PD. The lab is modeling these pathologies using patient-derived iPSC neuronal cultures and is testing novel spine regenerating agents in collaboration with a small biotech company to generate preclinical data sets for future clinical trials. These studies are accompanied with the development and use of a PET tracer to image spine loss and damage in cognitively impaired patients.

Dr. Elliott Mufson is an Institutional Professor in the Division of Neurobiology at Barrow Neurological Institute. Dr. Mufson is the director of several active grants, including a National Institute on Aging-supported Program Project grant entitled “Neurobiology of Mild Cognitive Impairment (MCI) in the Elderly” and a Department of Defense grant to study brain trauma. Dr. Mufson is a pioneer in the application of single cell gene array technology to study the genetic signature of neurons during the progression of AD. He has published 257 peer-reviewed articles and more than 40 book chapters. In 2010, the Information Sciences Institute recognized Dr. Mufson as one of the 100 most highly cited researchers in neuroscience

Dr. Fredric Manfredsson also recently joined BNI. His work focuses on the application of viral vectors in the study of and treatment of Parkinson’s disease and related neurodegenerative disorders. A significant portion of Dr. Manfredsson’s research program focuses on the protein alpha-synuclein and its role in disease and normal brain function. His lab has focused on engineering and characterizing recombinant Adeno-Associated Virus (rAAV) and Lentiviral vectors for the delivery to both the central and peripheral nervous system or cells in vitro. This includes the rational engineering of novel viral capsids and expression cassettes, or the use of molecular evolution to generate large AAV libraries. The Manfredsson laboratory then utilizes these engineered vectors to study, and treat, pathological molecular processes in neurodegenerative disease, such as Parkinson’s disease and Alzheimer’s disease.

# BARROW NEUROLOGICAL INSTITUTE at St. Joseph's Hospital and Medical Center Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on Project</b>
Burke, Anna	MD	Geriatric Psychiatrist
Wicklund, Meredith	MD	Neurologist
Baena, Elsa	PhD	Neuropsychologist
Elliott, Carol	BS	Manager, Biobank Core Facility
Neylon, Lizzi	BS	Coordinator, Biobank Core Facility
Mufson, Elliot	PhD	Neuroscientist
Wu, Jie	PhD	Neuroscientist
Perez, Sylvia E	PhD	Neuroscientist
He, Bin	BS	Research Technician
Garcia, Angelica	MS	Study Coordinator
Batchuluun, Dawn	BS	Program Administrator
Snell, Margeaux	MD	Study Coordinator
Hanson, Krista	PhD	Neuropsychologist
Vadovicky, Sheila	BS	Psychometrist
Martinez, Tiffany	BS	Psychometrist
Stokes, Ashley	PhD	MR Research, Keller Center for Imaging Innovation
Bergamino, Mauricio	PhD	MR Research, Keller Center for Imaging Innovation
Steffes, Lori	--	Study Coordinator
Sattler, Rita	PhD	Neuroscientist
Lorenzini, Ileana	PhD	Post-Doctoral Fellow
Moore, Stephen	PhD	Post-Doctoral Fellow

# CRITICAL PATH INSTITUTE

## Institutional Abstract

Critical Path Institute (C-Path) is a nonprofit, public-private partnership with the U.S. Food and Drug Administration (FDA) created under the auspices of the FDA's Critical Path Initiative program in 2005. C-Path's aim is to accelerate the pace and reduce the costs of medical product development through the creation of new data standards, measurement standards, and methods standards that aid in the scientific evaluation of the efficacy and safety of new therapies. These pre-competitive standards and approaches have been termed "drug development tools" (DDTs) by the FDA, which established a process for official review and confirmation of their validity for a given context of use. C-Path orchestrates the development of DDTs through an innovative, collaborative approach to the sharing of data and expertise. We build consensus among participating scientists from industry and academia with FDA participation and iterative feedback. The process culminates in a formal application to FDA for official "qualification" of the DDT for a given use in product development. Qualified DDTs then become open standards for the scientific community which, in turn, may be assured both of the scientific rigor under which they were developed and of the FDA's understanding and acceptance of their validity.

**The Critical Path for Alzheimer's Disease (CPAD) consortium** accelerates drug development for patients with chronic neurodegenerative disease leading to dementia, primarily Alzheimer disease, by advancing Drug Development Tools (DDTs) for evaluating drug efficacy and safety, working with industry and advocacy organizations to optimize novel clinical trial designs, and aggregating anonymized patient-level data using CDISC consensus standards to facilitate the regulatory review process.

CPAD is collaborating with industry, regulators, academia and philanthropic donors to leverage the wealth of drug development knowledge that the consortium members (industry members as well as academic researchers) possess, by enabling pre-competitive widespread data sharing from clinical trials in AD and contribute directly to the availability of new effective treatments for AD by focusing on the tools and knowledge needed to support successful drug development. By expanding CPAD's existing database and by enabling a rich clinical trial repository, CPAD will contribute directly to the generation of actionable solutions for drug development across the AD continuum. This database will drive the potential for scientific discovery provided by aggregated and standardized primary clinical trial data and resulting quantitative tools will, in turn, provide solutions to optimize the design of clinical trials of AD drugs intended for regulatory review in support of marketing approval.

# CRITICAL PATH INSTITUTE

## Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on Project</b>
Sivakumaran, Sudhir	PhD	PI, Executive Director, Critical Path for Alzheimer's Disease
Burton, Jackson	PhD	Mathematician
Hanan, Nathan	PharmD	
Karten, Yashmin	PhD	
Romero, Klaus	MD	

# MAYO CLINIC ARIZONA

## Institutional Abstract

The main goal of this research program is to determine the correlation between genetic risk for Alzheimer's disease (apolipoprotein E [APOE] genotype) and the effect of normal aging on certain measures of cognitive function, brain volume, brain metabolism, cerebral amyloid deposition, and potential plasma biomarkers (APOE fragments and others). The principal institutions involved in this collaborative research effort are Mayo Clinic Arizona (primary site), Banner Alzheimer's Institute, Barrow Neurological Institute, Arizona State University, and Translational Genomics Research Institute; though as the program has matured, it has evolved into a core resource for investigators from these and other institutions as well. Our research program capitalizes on the clinical and neuropsychological expertise of the Behavioral Neurologists and Neuropsychologists at Mayo Clinic Arizona, in conjunction with the genetic expertise of Drs. Eric Wieben, Eric Klee, and Rory Olson at Mayo Clinic Rochester and Dr. Matthew Baker at Mayo Clinic Jacksonville. This past year we were also fortunate to be joined by Dr. John Fryer, a key member of the neuroscience team at the Mayo Clinic in Jacksonville, Florida whose relocation to Arizona further strengthens our ties with the Florida neuroscience labs. Dr. Fryer's work has focused on the genomics of neurodegeneration including less well understood areas such as dark and camouflaged genes as well as amyloid independent pathways of neurodegeneration related to APOE, TREMs, and tau variants.

Our longitudinal study design is a unique strength with our longest participants having been followed for up to 25 years. Cognitive and related behavioral data are analyzed with regard to demographic and health related factors (e.g., hypertension), APOE genetic status, physical and psychological stressors, cognition, and brain imaging measures. We have shown the neuropsychologically defined onset of Alzheimer's disease begins during our 50's in APOE e4 carriers, is confined to memory during the early preclinical phase but there is an increase in self-awareness of decline that is not mirrored by informant observation. In later stages of preclinical Alzheimer's disease, as patients get within a few years of incident MCI conversion, executive measures begin to decline and informant observations begin to parallel self-reports of decline. Finally, by the time MCI emerges, memory, executive skills, and in some cases visuospatial skills begin to decline; and subtle personality changes begin characterized by increased proneness to stress and reduced openness to new ideas and experiences. Missing from the preclinical profile is any indication of depression, but the development of personality changes lays the groundwork for behavioral manifestations which begin to emerge during the MCI stage.

In addition to our cognitive studies, we have created a biobank of plasma, serum, and DNA that has served as a core resource for collaborative members.

To date we have:

1. Analyzed the longitudinal trajectories of all our measures, identified those showing significantly greater acceleration of decline in APOE e4 carriers relative to noncarriers, and developed a cognitive profile of APOE e4 driven pathological aging that defines the cognitive profile of preclinical Alzheimer's disease.
2. Compared our incident cases of mild cognitive impairment (MCI) to a clinical (prevalent) group of matched patients to further define an early and late preclinical/early clinical phase in which we begin to see decline in non-memory measures, especially those sensitive to executive functions.

3. Characterized the significance of subjective impairment as voiced by one's self as well as by one's informant and showed that both reflect an early stage of decline in a small subset, but that stress related symptoms overshadow the cognitive changes so that subjective impairment alone is an unreliable indicator of imminent decline.
4. Showed that personality traits that increase one's proneness to stress further speed up age-related memory decline, and this effect is more apparent in APOE e4 carriers reflecting their inherent predilection for Alzheimer's disease. In contrast, we found that the developmental sex-based cognitive advantages of women over men regarding verbal memory and men over women regarding visual memory do not buffer the rate of decline associated with APOE e4.
5. Presented an analysis of a computer-based cognitive task developed by Mario Parra sensitive to memory "binding" of different stimulus properties (e.g., shape and color), but we did not find this to be more sensitive than conventional neuropsychological measures of declarative memory.
6. Completed a survey, both online as well as among members of our cohort, examining attitudes about predictive testing for Alzheimer's disease (genetic and biomarker based) and found there is considerable interest in having such testing even in the absence of definitive therapy, but that roughly 12% and 6% respectively envision suicidal ideation should they be found at high risk for Alzheimer's disease. These results are informing the design of test disclosure methods in forthcoming trials.
7. Identified and characterized participants who have some behavioral features of a "broad autism phenotype" and showed how that influences subjective cognitive decline
8. Showed for the first time, actual personality changes that coincide with the transition from normal cognition to mild cognitive impairment and that in turn lay the groundwork for the behavioral disruptions that are prevalent in patients with MCI and dementia.
9. Showed that the earliest cognitive changes in individuals who eventually develop MCI and dementia begin roughly 20 years before incident MCI diagnosis, within the same general timeframe as the most sensitive biomarkers challenging current pathophysiological models.

These types of analyses will continue well into the future permitting us to achieve our longer-term goals of:

1. Correlating changes in brain function with structure, metabolism, and pathology including biomarkers in living patients.
2. Determining rates of symptomatic conversion from preclinical Alzheimer's disease to MCI, and from MCI to dementia.
3. Developing a predictive model based on presymptomatic parameters for the timing of symptomatic conversion.
4. Developing primary prevention strategies.
5. Providing a core resource to all our collaborative partners.
6. Correlating nontraditional measures of neuropsychiatric status such as intellectual achievement, sleep patterns, and personality factors with presymptomatic cerebral amyloid levels.

Specific goals for this fiscal year include:

1. Leveraging our cross sectional and longitudinally collected plasma and serum samples for a study of blood based biomarkers during the preclinical phase of AD.
2. Applying an epigenetic algorithm to brain derived DNA of nonfamilial young onset AD cases to determine whether such brains have an epigenetic signature of advanced biological age as an explanation for unusually young onset dementia.

3. Correlating our longitudinal imaging data with our now published neuropsychological data to contrast the chronology of MRI and FDG-PET changes with neuropsychological changes.
4. Exploring the development and application of supportive programs for our MCI and dementia patients and their caregivers.

This research proposal has been peer reviewed and approved by the Mayo Clinic Institutional Review Board (IRB #259-99).

# MAYO CLINIC ARIZONA

## Key Personnel

Name (last, first)	Degree	Role on Project
Caselli, Richard	MD	Principal Investigator, Clinical Core Director, Associate Director, Behavioral Neurologist
Woodruff, Bryan	MD	Co-Investigator, Behavioral Neurologist
Locke, Dona	PhD	Co-Investigator, Neuropsychologist
Stonnington, Cynthia	MD	Co-Investigator, Psychiatrist
Ezenne, Adaeze	NP	Nurse Practitioner
Henslin, Bruce	BA	Study Coordinator
Brostrom, Debra	BA	Study Coordinator
Baxter, Leslie	PhD	Neuroimaging Scientist
Fryer, John	PhD	Neuroscientist

# MIDWESTERN UNIVERSITY

## Institutional Abstract

Midwestern University (MWU) is a university of health sciences dedicated to the education of future health professionals. MWU has Colleges of Osteopathic Medicine, Graduate Studies, Optometry, Dental Medicine, Pharmacy, Veterinary Medicine, and Health Sciences. There are also 13 additional programs including new Precision Medicine and Master of Public Health Programs. We have multiple university-based clinics including the Multispecialty Clinic, the Eye Institute, the Dental Institute, and the Companion Animal Clinic. MWU has a rapidly growing and diverse research community focused on disease-specific research as well as basic science research. Our scientists and clinicians (both human and veterinary) are involved in many different research efforts, with collaborations throughout Arizona and the U.S. MWU supports a broad range of research, from neurological disorders and cancer to infectious diseases and anatomical studies. The research environment at MWU is highly collaborative and designed to use the collective expertise of our colleagues to achieve common goals.

Multiple interdisciplinary research programs have been developed in the last few years and are thriving. The MWU Institute for Healthcare Innovation (IHI) provides a comprehensive setting to conduct clinical trials, translational research and technology development regarding human and veterinary drugs, biologics, devices, nutritional products, and diagnostics. Midwestern has also developed the Nanomedicine Center of Excellence in Translational Cancer Research, with the goal of applying new technologies to the treatment of cancer. Our Veterinary Medicine program has brought with it many new research opportunities which support the Midwestern University One Health Initiative, which focuses on bringing together both basic and clinical researchers from our various colleges to gain insights into the interrelationships between public health, biodiversity and sustainability. Our goal is to train our students in the interdependence of all healthcare professions, for the benefit of current and future patients.

To support the goals of the Arizona Alzheimer's Consortium, the faculty at Midwestern University have created a formal group, the Midwestern Alzheimer's Advisory Committee (MAAC), dedicated to research into Alzheimer's disease (AD) and related conditions. This group now includes faculty from 16 departments/programs and multiple colleges. The goals of MAAC are to 1) leverage this diversity of expertise and establish a common core of investigators that contribute to our understanding of neurodegenerative disorders and aging, 2) inspire collaboration within MWU and with investigators at other institutions, and 3) complement and enhance the efforts of other Consortium-affiliated institutions and investigators around the state. Future goals for Midwestern University's Consortium efforts include broader roles in basic science understanding, patient evaluation and treatment mechanisms, education and outreach, and clinical recruitment.

Current Alzheimer's research-related activities at Midwestern include:

- 1) Understanding the potential role of microbes in the development of Alzheimer's disease brain pathology and cognitive deficits. This research involves studies of 1) human post-mortem tissues, including patients with both AD and MCI in comparison to normal and high pathology non-demented controls, 2) cell culture models of neuronal infection with microbes previously identified as being present in AD patients, and 3) infection of 3xTG

and APOE4 mice to test if infection with common microbes can exacerbate pathology in these models.

- 2) Determining the ability of genistein and exercise to 1) reverse inflammatory state, 2) modify brain protein expression, 3) modify gut leakiness, 4) modify microbiome, and 5) improve bone health in mice fed a high-fat diet (HFD). The goal of this project is to examine the link between metabolic syndrome and dementia, and test a drug which may be useful for modifying the cognitive outcome in patients.
- 3) Developing and validating new pharmacological treatments, such as norclozapine, that could have a positive impact on Alzheimer's disease and other neurological conditions, and support research on the cellular- and subcellular-targeted delivery of relevant treatments.
- 4) Evaluating the dysfunction within and contribution of various neurotransmitter systems in Alzheimer's disease and related disorders, such as Parkinson's disease, prominently including the nicotinic and muscarinic receptor systems of the brain.
- 5) Examining a proposed link between a protein that protects the chromosome ends against shortening (RAP1) and a protein localized to astrocytes (GFAP $\delta$ ), which also interacts with presenilin-1. Telomere shortening is a molecular cause of cellular aging, and advancing age is the greatest known risk factor for AD. This project studies the possibility that GFAP $\delta$  variants will modulate the accumulation of amyloid deposits in a cell culture model.
- 6) Examining the involvement of inflammatory molecules in the pathophysiology of Alzheimer's disease, related disorders, and CNS injury.
- 7) Determining whether elevated APOE4 expression is linked to cerebrovascular dysfunction in young and aged APOE4 mice, by measuring middle cerebral artery (MCA) function in APOE3 and APOE4 mice.
- 8) Studying exercise-induced mitigation of cellular senescence as a peripheral control mechanism for Alzheimer's disease using a senescence-accelerated SAMP8 mouse model.

# MIDWESTERN UNIVERSITY

## Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on project</b>
Jentarra, Garilyn	PhD	Administrative PI, Project Principal Investigator
Al-Nakkash, Layla	PhD	Principal Investigator
Anderson, Sarah	MOT/OTR	MAAC Investigator
Bae, Nancy	PhD	Principal Investigator
Broderick, Thomas	PhD	Principal Investigator
Castro, Monica	BS	Senior Research Associate
Christensen, Stephanie	PhD	MAAC Investigator
Chu, Ping	BS	Research Associate
Delgado Flint, Melissa	PsyD	MAAC Investigator
Eckman, Delrae	PhD	Principal Investigator
Esfandiarei, Mitra	PhD	MAAC Investigator
Fitzgerald, Nancy	DDS	MAAC Investigator
Gallas, Genna	MS	Research Associate
Gonzalez, Fernando	PhD	Co-Investigator
Halket, Christine	DDS	MAAC Investigator
Hernandez, Jose	PhD	MAAC Investigator
Huang, Vanthida	PharmD	Co-Investigator
Jadavji, Nafisa	PhD	MAAC Investigator
Jones, Carleton	PhD	MAAC Investigator
Jones, Douglas	PhD	Co-Investigator
Jones, T. Bucky	PhD	Principal Investigator
Kaufman, Jason	PhD	Co-Investigator
Kingston, Shanika	BS	Research Associate
Knudsen Gerber, Dawn	PharmD	MAAC Investigator
Kokjohn, Tyler	PhD	MAAC Investigator
Kozlowski, Michael	OD, PhD	MAAC Investigator
Lawson, Kathy	PhD	Co-Investigator
Li, Weidang	PhD	Co-Investigator
Murthy, Ashlesh	PhD	Consultant
Olsen, Mark	PhD	MAAC Investigator
Potter, Pamela	PhD	Principal Investigator
Potter, Ross	PhD	Laboratory Manager
Powell, Jessica	PsyD	Co-Investigator
Ratiu, Ileana	PhD	MAAC Investigator
Revill, Ann	PhD	MAAC Investigator
Rogers, Alexandra	BS	Research Associate
Shim, Minsub	PhD	MAAC Investigator
Swanson, Mark	PhD	Principal Investigator
Tullot, Tony	MD	Co-Investigator
Turner, Tamara	EdD, OTR	MAAC Investigator
Vallejo-Elias, Johana	PhD	Co-Investigator
Veltri, Charles	PhD	MAAC Investigator
Yevseyenkov, Vladimir	OD, PhD	MAAC Investigator

# NORTHERN ARIZONA UNIVERSITY

## Institutional Abstract

The Pathogen and Microbiome Institute (PMI) is based at Northern Arizona University (NAU). NAU ranks in the top 10 among all four-year, public institutions in Native American graduate student enrollment and in the top 100 of the National Science Foundation's research university ranking for research activity. The Center for Applied Microbiome Science at the Pathogen and Microbiome Institute has begun to engage in research on establishing a link between Alzheimer's Disease (AD) progression and the gut microbiota (the collection of microorganisms that inhabit an individual's gastrointestinal (GI) tract). To do this, we have established colonies of multiple murine models of single and triple transgenic AD and corresponding wild-type mice for analysis of the GI microbiome and AD-associated pathology throughout the course of AD progression.

To accomplish our research goals, we leverage our AAALAC-certified animal facility, a state-of-the-art BSL-2+ laboratory, and a large capacity for sequencing and computing power to complete cutting edge studies of the microbiota in Alzheimer's disease. NAU hosts a high performance computing cluster ("Monsoon") that has all of the software needed for microbiome and transcriptome analyses installed, including the popular QIIME 2 microbiome bioinformatics platform (<https://qiime2.org>; developed by PI Caporaso's team of students and professional software engineers at PMI). NAU and TGen North, located approximately one mile apart, share a sequencing core comprising three Illumina MiSeq machines, an Illumina MiniSeq, an Illumina NextSeq, and a MinION (Oxford Nanopore). The Joint Sequencing Core provides easily accessible sequencing for all faculty and staff at PMI, by following specific systems for sample tracking, preparation, and output data transfer. The core also serves as a resource in the dissemination of novel methods and provides training for new staff in sample preparation.

The goals of our research in the AAC are to assess changes in microbiome composition in the gut and other body sites that correlate with AD disease progression. We hope that these studies will lead to microbiome-based diagnostics or predictors of AD that can be used to delay or prevent the onset of this devastating diagnosis. In our current and future studies, we aim to establish a causative relationship between microbial community members and AD pathology and to translate findings from a preclinical murine model to human disease.

Our team at Northern Arizona University is well-positioned to achieve these goals. Dr. Cope has extensive experience with transcriptome analysis and microbiome research, and Dr. Caporaso is an expert in microbiome analysis, including recent work on using fecal microbiota transplant to improve behavioral symptoms of autism in a Phase 1 clinical trial, and on exploring the potential of features of the human oral microbiota for early cancer detection. In addition to our laboratory and sequencing capacity, we are developing laboratory and bioinformatics best practices for microbiome research. This includes automated nucleic extraction methods, application and validation of the latest microbiome sequencing protocols, and development of QIIME 2 (led by PI Caporaso), a microbiome bioinformatics platform. Key features of QIIME 2 is specifically designed for analysis of the type of data being generated in this project, and the platform is focused on ensuring reproducibility and transparency of microbiome analysis. We are therefore uniquely positioned to advance knowledge of the relationship between the gut microbiota and AD. These goals are achieved through decentralized data provenance tracking wherein each step of the analysis is automatically recorded and easily obtained in the results.

# NORTHERN ARIZONA UNIVERSITY

## Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on Project</b>
Cope, Emily	PhD	PI and Project Director
Caporaso, J Gregory	PhD	PI
Keim, Paul	PhD	Executive Director, PMI
Bolyen, Evan	BS	Research Software Engineer
Borsom, Emily	BS	Graduate Student
Conn, Kathryn	--	Undergraduate Researcher
Dillon, Matthew	MS	Research Software Engineer
Hirsch, Allyson	--	Undergraduate Researcher
Jaramillo, Sierra	BS	Graduate Student
Keefe, Chris	--	Student Research Software Engineer
Lee, Keehoon	PhD	Postdoctoral Scholar
Naimey, Turan	--	Student Research Software Engineer
Orisini, Gabrielle	--	Undergraduate Researcher

# **TRANSLATIONAL GENOMICS RESEARCH INSTITUTE Institutional Abstract**

The Translational Genomics Research Institute (TGen) is a non-profit biomedical research institute whose mission is to make and translate genomic discoveries into advances in human health. TGen is dedicated to bringing the breakthroughs in genomics research to the bedside and benefit of patients. Its focus on translational research involves coupling, in novel ways, basic and clinical science with emerging molecular technologies to accelerate the development of therapeutics and diagnostics for human disease. Part of the unique nature of TGen is its collaborative relationships with academic institutions, clinical practices and corporate entities, each aimed at accelerating discovery-based research towards application.

The Neurogenomics Division of TGen is the home of Alzheimer's disease (AD) and aging research programs within TGen. AD and aging has been a focus of the Division since its inception. The Neurogenomics Division is subdivided into several disease-oriented research clusters. Each cluster represents a unique cross-pollination between basic researchers and clinicians with the endgame being successful clinical trials that ultimately lead to improved treatments and diagnosis. These clusters include geneticists, molecular and cellular biologists, brain imaging researchers, proteomics specialists, drug development teams, and other experts.

The Division has accomplished several milestones in AD research including: (1) the first high-density genome screen to identify common heritable risk factors for AD, (2) the identification of a key genetic driver of episodic memory function in healthy individuals, (3) the first large-scale study identifying cell-specific genes differentially expressed in pathology-containing and pathology-free neurons in the brains of AD patients and control donors, (4) the identification of protein kinase targets responsible for phosphorylation of the tau protein which contributes to AD pathology and the use of this information to identify novel therapeutic approaches to the disease, (5) the collaborative discovery of a novel cognitive enhancing agent based on the genetic finding in episodic memory, and (6) the identification of new, cell-free extracellular vesicle biomarkers in the blood of AD patients. Collaborations within Arizona and across the nation have been critical for each of these projects and they include work with Arizona State University, Banner Alzheimer's Institute, University of Arizona, Banner Sun Health Research Institute, Barrow Neurological Institute, the National Institutes of Health, and many others.

Currently the Division has major areas of focus in the genetic basis of disease in rare AD clinical cases (using next generation DNA sequencing), the characterization of the transcriptome of multiple cell types in the AD brain (using laser capture microdissection and single cell sequencing approaches), cell-free fluid biomarker identification (using extracellular vesicle molecular profiling), and novel drug development for cognitive enhancement and AD. The Division also serves as an AD-related genomics and biostatistics resource for the Arizona Alzheimer's Consortium and frequently assists in generation and interpretation of genotyping and sequencing data.

Overall, the mission of the Division's work in AD is to develop improved ways to assess personalized risk for AD before the onset of symptoms, leverage molecular information to identify novel drug targets, and gain deeper understanding of the genomic changes associated with disease onset and progression.

# TRANSLATIONAL GENOMICS RESEARCH INSTITUTE

## Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on Project</b>
Alsop, Eric	PhD	Bioinformatician
Antone, Jerry	BS	Research Associate
Bonfitto, Anna	MS	Research Associate
DeBoth, Matthew	BS	Bioinformatician
Elyaderani, Amir	BS	Bioinformatician
Enriquez, Daniel	BS	Bioinformatician
Henderson-Smith, Adrienne	PhD	Research Associate
Henson, Sierra	BS	Research Associate
Hutchins, Elizabeth	PhD	Computational Scientist
Huentelman, Matthew	PhD	Principal Investigator
Jepsen, Wayne	MS	Graduate Student
Lechuga, Cynthia	MBA	Sr. Grants & Contract Administrator
Lewis, Candace	PhD	Postdoctoral Fellow
Liang, Winnie	PhD	Co-Investigator
Meechovet, Bessie	BS, BSN	Research Associate
Naymik, Marcus	MS	Bioinformatician
Piras, Ignazio	PhD	Research Assistant Professor
Reiman, Eric	MD	Consultant
Reiman, Rebecca	BA	Research Associate
Robles, Laura	MBA	Project Accountant
Talboom, Joshua	PhD	Postdoctoral Fellow
Van Keuren-Jensen, Kendall	PhD	Co-Investigator

# UNIVERSITY OF ARIZONA

## Institutional Abstract

Researchers at the University of Arizona (UA) are engaged in collaborative, multi-disciplinary programs of research focused on advancing our understanding of the major risk factors for brain aging and age-related neurodegenerative disease, their underlying neural substrates, and ways to prevent, delay, or treat cognitive aging and dementia. To accomplish these goals, UA investigators representing 12 departments and institutes, including researchers in the fields of neuroimaging, cognitive and behavioral neuroscience, neuropsychology, psychiatry, neurology, pharmacology, and statistical analysis are involved in these research programs. Projects apply a range of scientific approaches, from basic neuroscience to cognitive science to clinical intervention in studies that translate across species with humans and non-human animal models of aging and age-related disease. A major component of this research uses magnetic resonance imaging (MRI) as a cross-cutting method to measure brain function, structure, and connectivity in aging and age-related, neurodegenerative disease.

UA's researchers engage in translational research that spans multiple areas of expertise and methods to address clinical and basic research concerning the effects of healthy and pathological aging, including 1) investigating the neural systems and associated cognitive processes that are altered in the context of aging and age-related disease, 2) tracking brain changes and cognitive abilities during aging, 3) evaluating how genetic, health, and lifestyle factors affect brain aging and cognitive decline, 4) developing new behavioral and neuroimaging methods to improve early detection and associated brain changes due to aging and disease, 5) understanding cellular mechanisms of brain aging in animal models, 6) identifying and testing novel therapeutic and other interventions to improve cognitive function during aging, and 7) creating libraries and repositories for data sharing.

Over the past year, we have continued to establish the resources required to build a database for sharing standardized measurements that will be made available to all AAC researchers, utilizing XNAT, a shared online repository for neuroimaging data that is funded by the NIH. The complexity and high cost of collecting large-scale datasets highlights the importance of sharing data across laboratories. The database includes neuropsychological, neuroimaging, and biospecimen data obtained from well-characterized older adults. Standardized pipelines for MRI data analysis and establish standardized protocols for collection of biomarkers from blood and CSF have also been successfully established.

Program-related activities at the UA include several major areas of research:

**Neuroimaging development and application.** Our researchers continue to develop and implement new MRI techniques and statistical analysis methods that may prove useful in examining brain structure, function, connectivity, and pathology in both human and non-human animal models of aging and age-related disease. MRI methods including high-resolution structural imaging, fMRI, diffusion, perfusion, and resting state connectivity are being utilized to better understand the neural basis of memory and other cognitive changes across the normal adult lifespan, and compensatory or adaptive strategies that lead to better memory function. New technologies and methods are also being explored, including the use of MRI-guided transcranial magnetic stimulation to ameliorate memory impairment in patients with MCI, improving methods for imaging and measuring volumes of subregions of the thalamus, and ways to quantify and combine information from CBF and BOLD signal in MRI to better characterize the aging brain.

**Early detection and risk factors for AD.** A major theme of our research continues to focus on the early detection, diagnosis, and tracking of cognitive and psychological impairments associated with aging and Alzheimer's disease (AD). Several novel targets include subtle memory changes associated with hippocampal and perirhinal cortical functions, disturbances in patterns of daily thought, rumination, sleep disruptions, exercise history, and preclinical changes in MRI connectivity that may signal the effects of AD pathology prior to the onset of significant cognitive symptoms and changes in activities of daily living. Multiple projects focus on identifying and understanding the factors that increase risk for age-related cognitive impairment and AD, including physical activity, sleep quality, the quality of social interaction, as well as health factors such as hypertension, head injury, and cardiovascular fitness.

**Neural mechanisms and interventions.** Research projects are studying various potential targets for intervention. These include the potential neuroprotective effect of MAS antagonists, cellular mechanisms of vasculopathy, and the expression of melanocortin receptors. Each study has the potential to lead to novel interventions that may decrease risk for AD, slow the progression of the disease, and ameliorate cognitive impairments associated with normal aging and AD. These interventions include behavioral interventions such as exercise and cognitive engagement, transcranial repetitive stimulation, as well as novel pharmacological interventions including minocycline.

This program of research is complemented by our close ties to other research units at UA including the Evelyn F. McKnight Brain Institute, studying the longitudinal effects of aging on memory processes in older adults with and without increased risk for AD, and the Center for Innovation in Brain Sciences with a focus on the development of pharmacological interventions for degenerative brain diseases. UA researchers participate in complementary efforts to support the Arizona ADC with recruitment and longitudinal follow up of individuals with mild cognitive impairment, AD, and other forms of dementia. Our researchers are actively engaged in education and outreach in the Tucson community, partnering with a Diversity Outreach Program to enhance community outreach, education, and research participation by underserved minority groups in Arizona.

# UNIVERSITY OF ARIZONA

## Key Personnel

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on project</b>
Ahern, Geoffrey	MD	Investigator; Neurology, Psychology, Psychiatry, Evelyn F. McKnight Brain Institute
Alexander, Gene	PhD	Investigator; Psychology, Psychiatry, Neuroscience, Evelyn F. McKnight Brain Institute
Andrews-Hanna, Jessica	PhD	Investigator, Psychology and Evelyn F. McKnight Brain Institute
Arce, Fernando Teran	PhD	Investigator, Medicine
Barnes, Carol	PhD	Investigator; Psychology, Neurology, Neuroscience, Evelyn F. McKnight Brain Institute
Brinton, Robbie	PhD	Investigator, Center for Innovation in Brain Science, Pharmacology, Neurology, Psychology, Evelyn F. McKnight Brain Institute
Chen, Nan-Kuei	PhD	Investigator, Biomedical Engineering
Chou, Ying-hui	PhD	Investigator, Psychology and Evelyn F. McKnight Brain Institute
Edgin, Jamie	PhD	Investigator; Psychology
Ekstrom, Arne	PhD	Investigator, Psychology and Evelyn F. McKnight Brain Institute Investigator
Erickson, Robert	MD	Investigator, Pediatrics
Fernandez, Fabian	PhD	Investigator; Psychology, Neurology, Psychology, Evelyn F. McKnight Brain Institute
Gaffney, Kevin	PhD	Investigator, Pharmacology
Glisky, Elizabeth	PhD	Investigator; Psychology, Evelyn F. McKnight Brain Institute
Grilli, Matthew	PhD	Investigator, Psychology and Evelyn F. McKnight Brain Institute
Guzmán- Pérez Carrillo, Gloria	PhD	Investigator, Biomedical Engineers
Hay, Meredith	PhD	Investigator; Physiology, Psychology, Evelyn F. McKnight Brain Institute
Hishaw, G. Alex	MD	Investigator; Neurology, Psychiatry
Khanna, May	PhD	Investigator, Center for Innovation in Brain Science
Klimentidis, Yann	PhD	Investigator, Epidemiology and Biostatistics
Koshy, Anita	MD	Investigator; Neurology, Immunobiology, Evelyn F. McKnight Brain Institute
Matsunaga, Terry	PhD	Investigator, Medical Imaging
Mehl, Matthias	PhD	Investigator, Psychology and Evelyn F. McKnight Brain Institute
Nikolich-Zurich, Janko	PhD	Investigator, Immunobiology, Arizona Center on Aging
Raichlen, David	PhD	Investigator; Anthropology

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on project</b>
Rapcsak, Steven	MD	Investigator; Neurology, Psychology, Speech/Language and Hearing, Evelyn F. McKnight Brain Institute
Rodgers, Kathleen	PhD	Investigator, Center for Innovation in Brain Science
Ryan, Lee	PhD	Investigator; Psychology, Neurology, Neuroscience Program, Evelyn F. McKnight Brain Institute
Saranathan, Manojkumar	PhD	Investigator, Medical Imaging
Su, Judith	PhD	Investigator, Optical Sciences, Chemistry and Biochemistry
Trouard, Theodore	PhD	Investigator; Biomedical Engineering, Medical Imaging, Evelyn F. McKnight Brain Institute
Watts, George	PhD	Investigator, Pharmacology, Cancer Center
Weinkauf, Craig	MD,PhD	Investigator, Surgery
Wilson, Robert	PhD	Investigator, Psychology
Yin, Fei	PhD	Investigator, Center for Innovation in Brain Science
Zarnescu, Daniela	PhD	Investigator, Molecular & Cellular Biology, Neuroscience, Neurology
Zhou, Wei	MD	Investigator, Vascular Surgery

# **UNIVERSITY OF ARIZONA**

## **COLLEGE OF MEDICINE – PHOENIX**

### **Institutional Abstract**

The University of Arizona (UA) has a strong history of academic and medical excellence in the state of Arizona, governed by the Arizona Board of Regents. Two medical school campuses have been established, one located in Tucson at the Arizona Health Sciences Center and University Medical Center, and one located in Phoenix on the Phoenix Biomedical Campus (PBC). The UA College of Medicine – Phoenix shares the PBC campus with the UA College of Pharmacy, UA College of Public Health, UA Eller College of Management, and several allied health programs from Northern Arizona University, Arizona State University, and the Translational Genomics Research Institute. Through these many colleges and institutes, the UA College of Medicine – Phoenix is uniquely positioned to accelerate the biomedical and economic engines in Phoenix and the State by leveraging vital relationships with key clinical and community partners.

The UA College of Medicine – Phoenix mission is to inspire and train exemplary physicians, scientists, and leaders to optimize health and healthcare in Arizona and beyond. The UA College of Medicine – Phoenix was founded in 2007 as a full, four-year medical program. It was granted full independent accreditation by the Liaison Committee of Medical Education (LCME) in June 2017. At its new class size, the program matriculates 100 new allopathic doctors each year, with a total goal of 120 students per class. The UA College of Medicine – Phoenix continues to expand and grow as it also provides graduate training opportunities through the Clinical Translation Science Program. This program offers MS and PhD and combined MD/PhD and MD/MPH degrees.

The UA College of Medicine – Phoenix commits to life-long learning and critical thinking for all trainees, staff, and faculty. One example of this commitment is the requirement for all medical students to complete a Scholarly Research Project over their four years of medical training. Students are paired by the university with physicians and translational scientists to complete projects that cumulate in a thesis as part of the graduation requirements.

As part of the overall mission of the university, UA College of Medicine – Phoenix has developed and continues to reinforce cooperative agreements, partnerships, and collaborations with local institutions. Some examples include the development of the Translational Neurotrauma Research Program, a collaboration between the UA College of Medicine – Phoenix, Barrow Neurological Institute at Phoenix Children’s Hospital, and the Phoenix VA Health Care System. The Translational Neurotrauma Research Program sets the goal to be the premiere destination for neurotrauma research, training, and collaboration. The program has attracted scientist trainees and physicians from multiple world-renowned institutes and will continue to grow and prosper under these strong collaborations. More recently, this program has engaged with partners from the Maricopa County Attorney’s Office, Mesa Police Department, Tempe Police Department, HonorHealth Family Advocacy Center, Sojourner Center, and the CACTIS Foundation to establish the Maricopa County Collaboration on Concussion in Domestic Violence (MC3DV). Primary research directions for the program include inflammation, rehabilitation, and practical therapies for traumatic brain injury as a causative factor in challenging healthy aging and promoting neurodegenerative disease.

**UNIVERSITY OF ARIZONA  
COLLEGE OF MEDICINE – PHOENIX  
Key Personnel**

<b>Name (last, first)</b>	<b>Degree</b>	<b>Role on project</b>
Christie, Immaculate	--	Undergraduate research assistant
Giordano, Katherine R.	BS	Graduate student
Griffiths, Daniel R.	BS	Research Specialist, Senior
Hur, Yerin	BS	Technician
Law, L. Matthew	PhD	Co-investigator, Lecturer
Lifshitz, Jonathan	PhD	PI, Professor, Director
Rojas, M. Luisa	MS	Graduate student
Rowe, Rachel K.	PhD	Assistant Professor, Collaborator
Saber, Maha	PhD	Co-investigator, Post-doctoral fellow
Tallent, Bret R.	LATG	Laboratory manager
Young, Conor	BS	Technician

# **Project Progress Reports**

**Project Progress Reports**  
**Arizona State University**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Behavioral impact of surgical menopause variants with aging.** Heather Bimonte-Nelson, PhD. Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

The specific aim of this project is to determine whether hysterectomy continues to impair memory when length of time since surgery is extended into middle- and old- age.

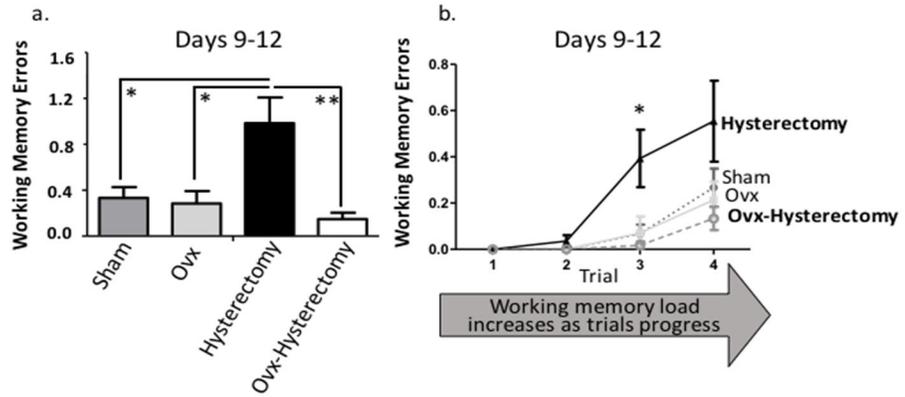
### **Background and Significance:**

Hysterectomy, or the surgical removal of the uterus, is one of the most common surgical interventions in women, second only to cesarean section (CDC, 2010; Carlson et al., 1993). Between 1998 and 2010, over 7.4 million women underwent hysterectomy, with approximately 600,000 surgeries occurring each year in the United States alone (Carlson et al., 1993; Corona et al., 2015, Wright et al., 2013). Of these surgeries, up to seventy percent of hysterectomies are performed in women under the age of 50, that is, before the average age of menopause (Wright et al., 2013). This surgical approach is most often taken to alleviate undesirable symptoms associated with benign uterine conditions. Gonadal hormones, particularly estrogens, have a well-documented effect on cognitive processes— especially those related to memory and affect.

Estrogens have long been considered to have neuroprotective properties, as well as beneficial effects on other body systems, such as bone and cardiovascular health. The sudden loss of ovarian hormones resulting from oophorectomy prior to natural menopause can be detrimental to memory in humans (Farrag et al., 2002; Nappi et al., 1999; Rocca et al., 2007, 2009, 2011, 2012) and in rodent models (Bimonte and Denenberg, 1999; Talboom et al., 2008; Wallace et al., 2006). *Yet, the surgical removal of the uterus alone does not result in the same drastic loss of circulating ovarian hormones that occurs with oophorectomy.* **Whether hysterectomy results in long-term effects on the brain and other body systems during aging remains to be fully explored.** In addition to research suggesting that hysterectomy prior to natural reproductive senescence can initiate ovarian failure earlier (Kaiser et al., 1989), Phung and colleagues reported an increased relative risk ratio to develop early onset dementia for women who underwent hysterectomy with and without oophorectomy compared to women with no history of hysterectomy; this increased risk was particularly evident when the women had a hysterectomy before the age of 30 years (Phung et al., 2010). Further, a greater risk of dementia arose when surgical menopause intervention (i.e., ovary removal with or without hysterectomy) occurred before the natural transition menopause (Rocca et al., 2007, 2012). Conversely, another research group found that Alzheimer's disease (AD) risk decreased with hysterectomy (with and without oophorectomy); however, the majority of women in this study were over 51, and therefore likely post-menopausal at the time of hysterectomy (Imtiaz et al., 2014). These incongruent findings suggest that, even with ovarian preservation, hysterectomy may alter the trajectory of brain aging in women and could be an important factor for understanding healthy cognitive aging, thus warranting further investigation. We have shown hysterectomy-induced impairments in young animals (Figure 1, Koebele et al., 2019). The current application will evaluate whether the effects of hysterectomy present also in old age.

### **Experimental Designs and Methods:**

Young and aged animals will be given Sham, Hysterectomy, or Ovx-Hysterectomy surgery and then behavior testing. Testing will include the spatial working and reference memory water radial-arm maze, the spatial reference memory Morris maze, the open field test to assess anxiety-like and locomotor activity, and the control visible platform task to confirm motoric and visual procedural components of a water escape maze task. After testing, blood serum will be evaluated for steroid hormones and feedback hormones including LH and FSH. Brain measures related to hormone profiles will also be evaluated and correlated with behavioral outcomes.



**Figure 1:** Working memory errors on the water-radial arm maze in young rats given sham, ovariectomy (Ovx), hysterectomy, or Ovx plus hysterectomy surgery. (a) During the latter half of testing, there was a main effect of Treatment, whereby young animals with a hysterectomy made more errors overall. (b) This impairment was particularly evident when working memory load was most taxed. \* $p < 0.05$ , \*\* $p < 0.01$  (Koebele et al., 2019)

### **Proposed One-Year and Long-Term Outcomes:**

Surgeries and behavior testing will be completed by the end of the one-year project period. We expect to be scoring, analyzing, and writing the data into manuscript form soon after this time period, as well as completing brain assessments to correlate with behavioral data. Expected deliverables in a more long-term context include a manuscript submitted within two years from study initiation, and a grant to study brain/behavior/aging/hormone relationships with hysterectomy.

### **Year End Progress Summary:**

To date, we have ordered the animals for this project, and have given hysterectomy and sham surgeries to the rats. We waited the proposed timeframe, and have completed the entire behavioral testing battery including working and reference memory assessments, anxiety-like assessments, and a control task. Behavioral data scoring evaluations are underway. We expect that the full behavioral data collection and analyses will be completed by mid-summer. Hormone and brain evaluations have yet to be performed, and will allow us to determine relationships between surgical menopause status, circulating hormone levels, cognition, and aging. We anticipate that the manuscript will be submitted by the end of fall of 2020.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Using hiPSC-models to establish a causative link between traumatic brain injury (TBI), neuroinflammation, and Alzheimer's disease.** David Brafman, PhD, Sarah Stabenfeldt, PhD. Arizona State University; Arizona Alzheimer's Consortium.

### **Project Description:**

#### **Background and Significance**

Although the majority of Alzheimer's disease (AD) cases are sporadic, multiple genetic and environmental factors have been associated with increased risk of AD onset and progression. As such, several epidemiological studies have revealed a strong relationship between traumatic brain injury (TBI) and an increased risk of AD<sup>1-5</sup>. In addition, investigations with AD rodent models revealed that TBI accelerates A $\beta$  deposition and tau abnormalities as well as the onset of impaired spatial learning<sup>6-7</sup>. Confirmatory analysis of postmortem tissue of long-term survivors of a single TBI revealed extensive diffuse A $\beta$  protein plaques, similar to those seen in advanced AD<sup>8-9</sup>. Along similar lines, intra-axonal accumulation of phosphorylated tau, another pathological hallmark of AD, has been documented in cortical tissue of patients that have experienced a single TBI<sup>9</sup>. Despite these connections, the underlying mechanisms by which TBI induces AD-related pathology, neuronal dysfunction, and cognitive decline are significantly understudied and have yet to be elucidated.

It has been suggested that TBI leads to a neuroinflammatory response highlighted by activated microglia, reactive astrocytes, and elevated expression of pro-inflammatory cytokines<sup>10-11</sup>. Along similar lines, many of the pathologies of AD are mediated through a similar inflammatory cascade<sup>12</sup>. While there is some correlative evidence demonstrating that the pro-inflammatory cytokines that are induced by TBI-related mechanical injury can enhance APP levels and amyloidogenic APP processing, a causative link between TBI, neuroinflammation, and Alzheimer's disease has not been established. In this proposal, we will use a highly accessible human induced pluripotent stem cell (hiPSC)-based mechanical injury model of TBI to dissect the relationship between TBI-induced inflammatory response and AD-related phenotypes.

**Specific Aims.** In this project, we will use our collective expertise in hiPSC culture, traumatic brain injury, and Alzheimer's disease modeling to accomplish the following specific aims:

**Specific Aim 1: Examine the relationship between mechanical injury, inflammatory response, and AD-related phenotypes using a hiPSC-based model of TBI.** Previously, our collaborator, Dr. Stabenfeldt, developed a shear-based model of mechanical injury<sup>14</sup>. In this aim, we will use this model with 3-D co-cultures of hiPSC-derived neurons, astrocytes, and microglia to identify precise relationships between well-defined biomechanical inputs (strain, strain rate), inflammatory response, and disease status. As such, we will be able to address the following questions: (1) What is the effect of mechanical injury on the inflammatory profile of astrocytes and microglia? (2) Is there a difference in mechanical injury-induced inflammatory response in cells generated from non-demented control (NDC) and AD hiPSCs?

**Specific Aim 2: Establish a causative relationship between mechanical injury, inflammatory response, and AD-related phenotypes.** In Aim 1, we will determine the inflammatory cytokines that are upregulated in response to mechanical injury in hiPSC-derived

cells. In this aim, we will use pharmacological and gene knockdown approaches to determine the direct relationship between these specific inflammatory modulators and the manifestation or augmentation of AD-related phenotypes.

**Preliminary Data.** The Brafman laboratory is well-versed in the manipulation of hPSCs and the development of protocols for their directed differentiation towards neuronal and astrocytic lineages<sup>15-18</sup>. Importantly, as it relates to this proposal, we have shown that these hiPSC-derived astrocytes display a robust inflammatory response in response to stimuli such as lipopolysaccharide (LPS). More recently, the Brafman laboratory has developed several methods to generate 3-D hiPSC-based cortical cultures that better mimic the architecture and composition of *in vivo* neural tissue. In addition, the Brafman laboratory has extensive experience in the generation of hiPSC lines from NDC and AD patients and routinely applies a variety of biochemical and cellular assays to measure AD-related phenotypes in neural cultures derived from these hiPSC lines.

The Stabenfeldt laboratory has developed and characterized several different *in vitro* TBI models, which afford the ability to control bulk deformation and loading rate while monitoring cell response. In particular, the Stabenfeldt laboratory has developed a well-characterized *in vitro* injury model in which shear deformation can be imparted to 3-D cultures<sup>14</sup>. Using this system, the Stabenfeldt laboratory has established robust injury paradigms that mimic several phenotypes of *in vivo* neural injury including acute cell death, cellular hypertrophy and astrogliosis, and neuron plasmalemma disruptions.

#### **Year End Progress Summary:**

**Validation of cell-shearing mechanical injury model of TBI using 3-D hiPSC-derived neuron-astrocyte co-cultures.** We have further refined our differentiation protocols to reproducibly generate populations of mature hiPSC-derived cortical neurons or astrocytes with high purity. Briefly, these optimized differentiation protocols led to the generation of cells which express high levels of canonical astrocytic markers as well as display properties characteristic of functionally mature cells including production of Apolipoprotein E (APOE), responsiveness to inflammatory stimuli such as lipopolysaccharide, ability to uptake amyloid- $\beta$  (A $\beta$ ), and appearance of robust slow-decaying calcium transients. In parallel, our optimized neuronal differentiation protocols resulted in neuronal populations that express high levels mature neuronal-, neurotransmitter-, and cortical-related markers. In addition, these neuronal cultures display functional abundant rapid spontaneous calcium spikes indicative of functional maturation and synaptogenesis. Finally, RNA-seq analysis confirms that these differentiation protocols result in distinct neuronal and astrocyte populations free from contaminated hNPC populations (Figure 1J-K). Moreover, we have developed protocols so that these cells can be dissociated into single cell populations and cryopreserved without loss of functionality. The ability to generate large batches of astrocytes and neurons that can be characterized, cryopreserved, and thawed directly into the 3-D cultures will greatly improve the reproducibility of experiments. To that end, we have optimized various parameters including Matrigel concentration, seeding density, and astrocyte:neuron ratio to generate reproducible 3-D co-cultures in the injury chambers (Figure 1L). Importantly, during prolonged maintenance these 3-D co-cultures demonstrated high levels of cell viability, expression of key markers of maturation (Figure 1M), and robust calcium transients (Figure 1P). These optimized 3-D co-cultures will serve as the basis for our injury studies to be performed throughout the project.

In addition, we have modified the design cell injury device to provide more precise and reproducible control over the strain magnitude and rate that is imposed on the 3-D co-cultures. We are currently employing an iterative approach to develop a paradigm that induces injury-

related phenotypes in the hiPSC-derived 3-D culture in a manner similar to that we have previously achieved with primary rat neural cells.

**Examine the effects of mechanical injury on AD-related phenotypes in hiPSC-derived neural cells.** We have modified several of our existing protocols so that they are compatible with our 3-D co-culture system. For example, we have modified our immunofluorescence protocol so that 3-D co-cultures can be fixed, stained, and imaged with confocal microscope. Along similar lines, we have developed new protocols so that we can measure calcium transients in living cells embedded within the 3-D co-cultures. Moreover, through optimization of dissociation protocols we are able to collect living cells from the 3-D co-cultures to perform gene expression analysis as well as ELISA assays. In turn, these modified protocols will now enable us to measure AD-related phenotypes in injured and uninjured cultures to establish relationships between TBI-like injury, neuroinflammation and AD-related phenotypes.

***Proposed One-Year and Long-term Outcomes.*** The research we have performed as part of this proposal will allow us to establish a relationship between mechanical injury, production of inflammatory cytokines, and the manifestation/augmentation of AD-related phenotypes. The preliminary data and models that have been generated as part of this proposal will allow us to apply for more comprehensive grants. Specifically, we will submit a NIH R01-level application for the June submission cycle as well as collaborative research grant in response to the 2020 CDMRP Peer Reviewed Alzheimer's RFP.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Piloting an Evidence-based Intervention for Those Living alone with Mild Cognitive Impairment.** David W. Coon, PhD, Dona Locke, PhD, Arizona State University; Mayo Clinic Arizona; Arizona Alzheimer's Consortium.

### **Specific Aims:**

To gather and review qualitative data from focus groups and focused interviews to develop and pilot a psychosocial intervention to improve and maintain quality of life for people living alone with Mild Cognitive Impairment.

### **Background and Significance:**

Approximately 15-20% of people age 65 and older have Mild Cognitive Impairment (MCI), a condition characterized by measurable changes in thinking abilities that are noticeable to both people with MCI and their family/friends. However, people with MCI can still carry out their everyday activities. A recent systematic review suggests that approximately 32% of people with MCI go on to develop Alzheimer's within 5 years (Ward, Tardiff, Dye, & Arrighi, 2013). Depression appears to be quite common among MCI patients (25% in community samples; 40% in clinical samples) (Ismail, Elbayoumi, & Fischer, 2017), and MCI patients have reported significantly lower psychological quality of life compared to their peers with normal cognitive functioning. Moreover, living alone with MCI appears to place these MCI patients at higher risk for poorer outcomes (Muangpaisan et al., 2008). To date, no evidence-based treatments have been identified that improve and maintain quality of life for people living alone with MCI.

### **Preliminary Data and Plan:**

The two investigators for the proposed project run intervention programs for individuals diagnosed with MCI and/or early-stage dementia. Early-stage Partners in Care (EPIC), led by Dr. Coon as a partnership with ASU and the Alzheimer's Association, is a program focused on patients with early-stage dementia and their care partners. This group dyadic intervention includes education and skill-training workshops designed to reduced stress, enhance well-being, and help manage challenges by hearing the patient's voice in terms of care values and future care preferences. The HABIT Healthy Action to Benefit Independence and Thinking program, led by Dr. Locke at Mayo Clinic, is a cognitive rehab and brain wellness intervention for patients with MCI and a program partner. HABIT aims to support functioning, improved quality of life, and strengthen partnerships. EPIC and HABIT can be seen as companion programs as each involves different types of interventions. The HABIT program involves: (1) cognitive rehabilitation (2) support group for both patient and partner (3) wellness classes (4) cognitive exercise and (5) yoga. However, neither program is designed to support MCI patients who do not have someone to be their partner (e.g., individuals living alone with MCI with no local family members). Using our experiences with EPIC and HABIT as a frame, we want to respond to local and federal partner requests (e.g., the Alzheimer's Association, local Area Agencies on Aging, and the U.S. Administration for Community Living) to develop an intervention program for this population. Focus group data collected last year and this year will be used to guide the pilot intervention development and delivery.

### **Proposed One-Year and Long-Term Outcomes:**

The proposed short-term outcomes were to conduct additional focus groups and focused interviews with people living alone with MCI and the providers who assist this population, based

on findings from previous work; conduct and review analyses on data from these focus groups and focused interviews; use these analyses to develop screening, interview, and intervention components for an intervention for people living alone with MCI; conduct and analyze the data from a small single arm pre-post feasibility and acceptability project implementing the intervention. In addition, the data analyses would yield both professional presentations at meetings like the Gerontological Society of America, the American Society on Aging or American Psychological Association as well as the submission of the pilot results to venues like *The Gerontologist (Practice Concepts Section)*, *the Clinical Gerontologist*, or *Dementia*. Subsequently, the PIs would submit either an R21 or an R01 in 2021 or 2022, depending on the pilot project's findings.

**Year-End Progress Summary:**

Focus groups with health and social services providers working with older adults with memory concerns identified key issues to address in the development of the intervention protocol (recruitment, screening, assessment, and intervention development). Findings raised the critical need to capture participants “upstream” indicating that very few individuals have a diagnosis of MCI and that it is challenging to distinguish the “worried well” from those with MCI and those with early-stage dementia. Providers, particularly social services providers in the field, need a best practice screening tool to help them provide referrals to health care providers who can determine the appropriate diagnoses. Some health care providers suggested revising the project's current recruitment and screening materials to capture seniors with “memory concerns” and add additional telephone screening steps (e.g., telephone versions of the MOCA in combination with the TICS based on the current scientific literature) to determine those with symptoms consistent with MCI. Logistical barriers for intervention participation were also raised ranging from transportation/driving concerns to scheduling conflicts for those still employed. All components of the HABIT and EPIC projects were viewed as valuable. Recruitment of MCI participants for the focus groups has proven more challenging and the emergence of COVID-19 has delayed any delivery of the intervention. The PIs are revising the IRB protocols to permit telephone and/or video conferencing focus groups for those with MCI to gather additional feedback on intervention alternatives including delivery through online platforms.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

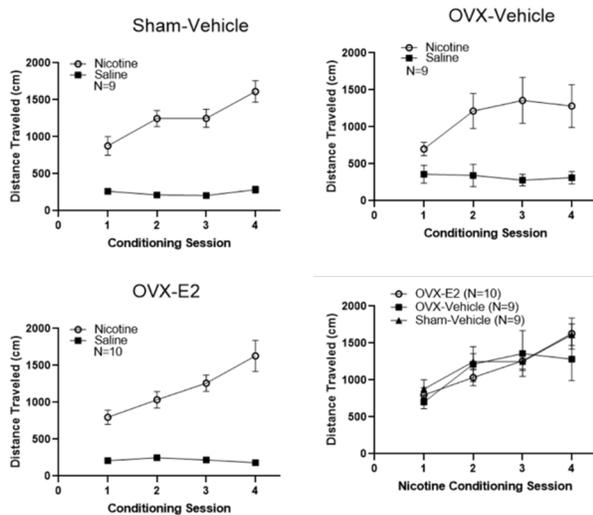
**An evaluation of neuroinflammatory processes in nicotine relapse during young adulthood and normal aging in females.** Cassandra Gipson-Reichardt, PhD, Heather Bimonte-Nelson, PhD. Arizona State University; Arizona Alzheimer's Consortium.

### Specific Aims:

To determine the role of NF- $\kappa$ B signaling in gating structural changes in synaptic plasticity during nicotine relapse in young adult females.

To determine if NF- $\kappa$ B activation drives nicotine seeking in aged females due to the loss of 17 $\beta$ -estradiol and the role of ER- $\beta$  receptors.

### Project Description and Progress Summary:



Women are typically more vulnerable to substance use disorders, and long-term smoking cessation is more difficult to achieve in women than men in clinical trials [1, 2]. The presence of menopausal symptoms is associated with lower rates of smoking abstinence [3]. As well, lifetime smoking is correlated with premature menopause [4], illustrating smoking as an important factor in reproductive health in aging women. Chronic nicotine use induces pro-inflammatory cytokine release [5, 6] and compromised cerebral blood flow [7]. Importantly, ovarian hormones interact with neuroinflammatory processes, including 17 $\beta$ -estradiol (E2). For example, E2 acts as an anti-inflammatory agent through inhibition of the nuclear factor-

$\kappa$  B (NF- $\kappa$ B) pathway [8]. Glutamatergic alterations within the nucleus accumbens core (NAcore) following withdrawal from nicotine self-administration render synapses of medium spiny neurons (MSNs) in a potentiated state [9-11], illustrating an important role of glutamate signaling in nicotine-motivated behavior. Further, Estrogens can activate NF- $\kappa$ B directly. Here we hypothesized that ovariectomized aged females given E2 will acquire nicotine conditioned place preference more readily, and that this is driven by interactions between E2 and the NF- $\kappa$ B pathway. Thus far, middle age (12 month old) and aged (18 month old) females have undergone nicotine conditioned place preference. Importantly, aged females show nicotine-induced enhanced locomotion with repeated nicotine administrations regardless of group (see figure above). These data are novel and indicate that acquisition of nicotine-related behaviors can happen across the lifespan, regardless of hormone status. CPP data analysis is underway, and we hypothesize that based on the locomotor sensitization data, all females have the necessary nicotinic receptors to sensitize dopaminergic signaling, and may show enhancement of nicotine CPP compared to animals not sensitized to nicotine. Brains from these animals have been extracted, and accumbens has been isolated for western blot. Throughout the duration of the award period, brain tissue will be processed for neuroimmune markers, including TNF $\alpha$ , p65, CD40, I $\kappa$ B, as well as GLT-1 and estrogen receptors (ER- $\alpha$  and ER- $\beta$ ).

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Are PSEN1 E280A mutation carriers an accelerated form of Dementia with Lewy body, not Alzheimer's disease?** Diego Mastroeni, PhD, Winnie Liang, PhD, Eric Reiman, MD, Ben Readhead, MBBS, Thomas Beach, MD, PhD, Ken Kosik, MD, Paul Coleman, PhD. Arizona State University; Translational Genomics Research Institute; Banner Alzheimer's Institute; Banner Sun Health Research Institute; University of California, Santa Barbara; Arizona Alzheimer's Consortium.

**Background:** As our population ages, neurodegenerative diseases including Alzheimer's disease (AD), and Lewy body dementia (DLB) (two most common form of dementia), will affect a significant and growing proportion of the population. In the US, over 5 million people currently suffer from AD, and 1.3 million with DLB, with an estimated cost for treating dementia of over \$230 billion dollars. A common thread linking all these neurodegenerative diseases is the presence of misfolded or alternatively folded proteins. Each disease has been primarily associated with aggregation of a specific protein or proteins—amyloid beta ( $A\beta$ ) and tau in AD, and alpha synuclein ( $\alpha$ -syn) and  $A\beta$  in dementia with Lewy bodies (DLB).

For nearly a century rare forms of AD known as early onset familial Alzheimer's disease (eFAD) have been described (reviewed in<sup>1</sup>). Harboring a mutation in the amyloid precursor protein (APP), presenilin-1 (PS1) or presenilin-2 (PS2) gene virtually guarantees that one will develop early onset AD. The E280A mutation in the PS1 gene is by far the most common mutation. The majority of E280A carriers belong to large kindred from the Colombian state of Antioquia. In these kindred, mutation carriers typically develop memory deficits in the third decade of life, followed by progressive impairments in other cognitive domains (reviewed in<sup>2</sup>). Researchers are looking for early pathological changes in presymptomatic E280A carriers using novel neuroimaging techniques, blood and CSF to gain knowledge of eFAD disease progression, but what are the pathological similarities and/or differences that distinguish AD from eFAD and DLB? One unique feature described in eFAD compared to late onset AD (LOAD) is the presence of Lewy body pathology (alpha synuclein aggregates), which is frequently found in the brains of eFAD patients harboring PSEN1 mutations<sup>2-4</sup>. Although overlapping neuropathological findings such as Lewy bodies have been described in LOAD this is largely considered "incidental", but is it?

**Overall Questions:** What are the overlapping biological pathways effected in individual neurons containing eosinophilic cytoplasmic inclusion (Lewy body) in LOAD with Lewy bodies, eFAD, and DLB? How do they differ from LOAD cases without mixed pathology, but do have "incidental" Lewy bodies?

**Hypothesis:** We believe that Lewy bodies are unique in their composition according to which underlying disease the patient presents neuropathologically. Although these cytoplasmic inclusions are identified using the same histopathological stains (e.g. haematoxylin and eosin) we hypothesize that 1) each disease will present unique expression profiles even though they have the same pathological hallmark (e.g. Lewy bodies), and 2) eFAD cases will resemble DLB cases in terms of expression profile more so than LOAD cases. To test this hypothesis, we will use a combination of immunohistochemistry and laser capture microdissection to select neurons containing Lewy bodies in eFAD, DLB and LOAD cases with Lewy bodies, and LOAD cases with "incidental" Lewy bodies. Captured cells will be processed for whole genome RNA sequencing, and multiple network modeling approaches will be performed to address the overlapping and unique expression profiles among neurodegenerative diseases with the same eosinophilic cytoplasmic inclusions.

## **Specific Aims:**

### **Aim 1: Capture individual neurons by LCM followed by RNA sequencing**

**1A)** We will capture immuno-positive Lewy body-containing neurons in eFAD, DLB, LOAD with LB and LOAD with “incidental” LB using established LCM methods<sup>5-9</sup>. 100 individual neurons containing Lewy bodies will be captured in the amygdala, an area of the brain that has been shown to have a dense population of Lewy bodies (Tom Beach). All subjects will be well matched (e.g. gender, age, LB stage, and postmortem intervals). We will use clinically and pathologically confirmed cases. All cases will be devoid of pathology other than that for their respective diseases.

**1B)** RNA processing and sequencing of LCM tissue, as previously described<sup>5-9</sup>.

**1C)** A list of differentiated genes will be compiled, and QPCR validation of the most significant genes will be analyzed as previously described<sup>5-7,10</sup>.

### **Aim 2: Network Modeling Approaches**

The purpose of Aim 2 is designed to address the commonalities and difference between diseases with the same neuropathology (e.g. Lewy bodies), but different clinical presentations. To demonstrate a relationship between these subjects, we will perform a differential analysis as implemented in the DESeq2 R-package as previously described<sup>5-7,10</sup>. Transcripts with corrected *P* value  $\leq 0.05$  and absolute log<sub>2</sub> fold change of  $>0.58$  or  $<-0.58$  (1.5 fold) will be considered differentially expressed. We propose that pathway analyses of differentially expressed transcripts will reveal commonalities and difference between groups. We hypothesize that eFAD cases will resemble DLB cases in terms of expression profile more so than LOAD cases.

**Innovation:** The innovation of this work largely derives by focusing on a specific class of cell, harboring the same cytoplasmic inclusion, in different neurological diseases, obtained by laser capture microdissection. Thus, the work proposed here will allow more precise definition of the effect of Lewy bodies in specific neurological conditions. These studies will have a major impact on defining disease not by the presence of Lewy bodies, but the effect Lewy bodies may have in different neurological conditions.

**Potential Pitfalls and Alternative Approaches:** 1) We recognize novel issues regarding quality control of RNA sequencing. For this reason, during library preparation, we will electrophoretically separate fragmented cDNA on a TAE gel to verify fragmentation. We will bioanalyze final libraries to verify sizes and quantitate libraries prior to sequencing. Finally, during and following sequencing, we will evaluate the total amount of Q30 data generated per library, total number of reads generated, and the total number of mappable reads. As with all sequencing technology there’s an inherent risk of false positives. For a subset of the identified altered transcripts, we will perform quantitative RT-PCR on all samples as previously described<sup>5-7,10</sup> 2) It is possible that we will not detect any substantial overlap in our comparison(s). This could conceivably occur as a result of: (a) not detecting any DE features, or (b) due to an actual lack of overlap. In the case of (a) we will also adopt a gene set enrichment analysis paradigm, which does not require significance-based thresholding of DE results, instead looking for transcriptome-wide shifts in groups of transcripts. This may allow us to detect a small but systematic shift in the group of known DE genes, even if no individual gene member was detected as DE. 3) The tissue from eFAD cases will be coming from Colombia. There is always an inherent risk when shipping samples from another country. Dr. Kosik recently obtained samples from Colombia so we know it’s possible, but what if there is a hold up? In this case we will continue analyzing DLB, LOAD with LB and LOAD with “incidental” Lewy bodies from our BSHRI brain bank. If the samples from Colombia take longer to arrive than anticipated, we will then use PS1 mutation carriers from our BSHRI brain bank. Currently there are 12 PS1 mutation carriers in the BSHRI brain bank.

**Future Goals:** The RNA sequencing data generated from these patients will be the first step in showing that the presence of Lewy bodies has differential effects on gene expression according

to the underlying diseases. The Future goal will be to identify the different molecules that make up Lewy bodies in different diseases by mass spectrometry.

**Year End Progress Summary:** All samples have been selected and secured. We thank Dr. Thomas Beach, Lucia Sue, Dr. Geidy Serrano and their staff for provision of tissue samples and postmortem evaluations. Laser capture studies are complete. We have cut and extracted RNA from all proposed cases except three. The three cases were excluded by Dr. Serrano. She thought she had enough PS1 cases, but it turned out that the samples were not of good enough quality for LCM studies. All samples will be ready for sequencing when the sequencing facility is re-opened.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Project Description

**Multiomic modelling of microbe-host interactions in the brain affected by late onset Alzheimer's disease.** Ben Readhead, MBBS, Winnie S. Liang, PhD, Thomas Beach, PhD, Diego Mastroeni, Christopher Mason PhD, Joel Dudley, PhD, Eric Reiman, MD. Arizona State University; Translational Genomics Research Institute; Icahn School of Medicine at Mount Sinai; Banner Sun Health Research Institute; Banner Alzheimer's Institute; Weill Cornell Medical College; Feil Family Brain and Mind Research Institute; Arizona Alzheimer's Consortium.

### **Project Description**

#### ***What are the major goals of the project?***

The goal of this project is to directly characterize the microbiome of post-mortem brain tissue in subjects affected by Alzheimer's disease (AD). We are performing shotgun metagenomic sequencing on post-mortem brain homogenate samples from subjects affected by AD, as well as normal controls (Total brain tissue samples: n=100, AD: n=50, Controls: n=50).

The data set being collected within this project is synergistic with two complementary data sets that we are collecting using NIH, and philanthropic funds. In combination, these three data sets will enable a detailed characterization of the microbial landscape of the brain, across the critical clinical and neuropathological inflections points of relevance to Alzheimer's disease (collectively: neuropathologically normal controls, preclinical AD, MCI, clinical AD, non-AD neurodegenerative disease). In each category, sample collection and profiling are proceeding in the same manner, maximizing comparability across groups.

#### ***What was accomplished under these goals?***

This project is currently at a stage that is characterized by logistical, rather than scientific tasks. This includes the prioritization and acquisition of tissue samples, establishing subawards with collaborators that will be performing sample processing, and optimization of protocols that will be used to prepare and process samples. We have made excellent progress with these steps. Although we have experienced some slow-down in sequencing as a result of COVID-related disruptions, we anticipate that data generation will commence in June/July of 2020, and accordingly the project will transition to a scientific focus.

#### **Brain and CSF Tissue requests from multiple biorepositories**

We are grateful to the Banner Sun Health Research Institute, who have fulfilled a request for 100 superior temporal gyrus brain tissue samples. The appropriate materials transfer agreements have been completed, and these samples have been delivered to Dr. Christopher Mason's lab (Weill-Cornell Medicine) to undergo sample processing and metagenomics sequencing.

#### **Shotgun metagenomics using a custom metagenomics capture panel**

To maximize our power to detect microbial sequences within tissue samples, we have chosen to perform an enrichment step that captures microbial sequences, prior to shotgun metagenomics sequencing step. This will greatly amplify our ability to detect relatively small (though potentially important) quantities of microbial sequences. This will be completed using a custom microbial sequence capture panel that has been designed in collaboration with Dr. Christopher Mason and is currently being manufactured by TWIST Biosciences. It is anticipated that this capture panel will be ready for use in May 2020.

**Expansion of project scope following cost reductions**

In combination with the 2 complementary projects referred to above, we are poised to profile approximately 3,000 brain tissue samples from a range of regions, and from subjects with a range of clinical diagnoses and endophenotypes. We are fortunate that due to the very large scope of these combined projects, the vendor that will be providing our capture enrichment panel (TWIST Biosciences) has substantially reduced their costs, allowing us to collectively profile three times the number of samples we have originally provisioned for. We are pleased to be now profiling a total of 300 samples as a result of this project, rather than the originally planned 100 samples.

***What do you plan to do for the next reporting period to accomplish the goals?***

We expect that over the next reporting period:

- (1) Custom TWIST microbial sequence capture panel will have been manufactured and shipped to the Mason Lab for incorporation into the sample processing.
- (2) Data generation will have commenced, and that we may be in a position to perform preliminary analyses on the available subset of data. Dependent on several factors, this will include comparative metagenomics between certain patient groups, brain regions, and institutions.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Injury-induced neuroinflammation as a contributor to Alzheimer's Disease.** Sarah E. Stabenfeldt, PhD, Salvatore Oddo, PhD, David Brafman, PhD. Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

**Specific Aim 1:** *Evaluate acute BBB dysfunction and neuroinflammation following traumatic brain injury in AD vs WT mice.* We will characterize BBB permeability and neuroinflammation in 3xTg-AD mice over the course of 7 days following a focal brain injury (CCI). The results from this study will provide unique insight into the contribution of AD risk factors on BBB dysfunction and inflammation following TBI and vice versa.

**Specific Aim 2:** *Evaluate chronic inflammation and markers of AD pathology following traumatic brain injury in AD and WT mice.* We will characterize neuroinflammatory and hallmark AD pathology in 3xTg-AD mice one month following a focal brain injury (CCI). The results from this study will provide unique insight into whether injury-induce neuroinflammation has a pronounced effect on AD pathology progression.

**Background and Significance:** An increased risk for Alzheimer's disease and Alzheimer's disease-related dementias (AD/ADRD) following documented TBIs has been identified in the clinic [1,2] and AD-like pathology has been observed in preclinical TBI models [3–5]. Commonalities exist between TBI and AD/ADRD pathologies including a dysfunctional blood-brain barrier (BBB) [6–8] and neuroinflammation [9–12]. Yet, the direct connection and potential contribution of TBI to AD/ADRD pathologies remains elusive. Notably, one hypothesis for AD/ADRD is the two hit vascular hypothesis postulating that an initial “insult” to the vascular system initiates BBB dysfunction to directly cause neuronal death. Continued BBB dysfunction then contributes to dysregulated clearance and subsequent deposition of amyloid- $\beta$  ( $A\beta$ ) leading to further neurodegeneration [7]. One could speculate that a TBI event could serve as the first vascular “insult” contributing to neuroinflammation and ultimately neurodegenerative sequelae. Therefore, elucidating key mechanisms following TBI, particularly relating to BBB dysfunction and neuroinflammation, that contribute to AD/ADRD will not only provide insight into basic pathological progression, but also spark novel therapeutic strategies to significantly impact both TBI and AD/ADRD. Here, we aim to first characterize the BBB permeability and acute neuroinflammation following TBI in the 3xTg-AD mouse model. Our analysis will focus on characterizing key aspects of the BBB dysfunction (tight junction, basement membrane, and  $A\beta$  receptors) and neuroinflammation (astrogliosis, microglial activation, cytokine profiles). Our second objective is to examine the influence of TBI on the chronic neuroinflammatory profile and AD pathology.

### **Preliminary Data, Experimental Design and Methods:**

**Preliminary Data:** Dr. Stabenfeldt is an established leader in evaluating BBB dysfunction following TBI in the proposed preclinical TBI model. She has recent publications that thoroughly characterized temporal BBB disruption profiles and also ability to exploit these windows of BBB disruption for localized nanoparticle deposition [13–15]. As an extension of this foundational work, Dr. Stabenfeldt recently received an NIH supplement on her DP2 NIH award to collaborate with Dr. Oddo and extend her work in TBI to the 3xTg-AD model. Through this funding (ends 6/30/19), we have conducted preliminary pilot studies to investigate initial BBB permeability and nanoparticle delivery in aged and 3xTg-AD mice following TBI. Moreover, Dr. Stabenfeldt's lab is well-versed in conducting IHC and proteomic analysis in mouse models [14–16]. Dr. Oddo is a

leading expert in the AD community, particularly in the proposed AD transgenic mouse model [17–19]. His group has extensive experience and data regarding the neuroinflammatory and AD pathology progression in the 3xTg-AD model.

### ***Experimental Designs and Methods***

**Specific Aim 1:** Cohorts of 3xTg-AD mice (female only at 20 weeks of age) and aged-matched WT controls will sustain a unilateral CCI followed by systemic intravenous injections of BBB permeability markers 2hrs prior to sacrifice at 3hr, 24hr, 3 day, or 7 days post injury. At each timepoint, tissue will be processed to evaluate BBB permeability, inflammation, and A $\beta$  deposition using proteomic and immunohistochemistry analyses. Immunostaining for inflammatory markers will include the following markers: Iba-1 (microglial/macrophage) and GFAP (astrocyte). BBB markers will include occludin and claudin-5. A $\beta$  deposition markers include A $\beta$  6E10 and 4G8, A $\beta$  1560, A11, and APP 22C11. Cytokine levels will be assessed via Bio-plex Cytokine Analysis with subsequent single cytokine ELISA follow-up as needed.

**Specific Aim 2:** Cohorts of 3xTg-AD mice (female only at 20 weeks of age) and aged-matched WT controls will sustain a unilateral CCI. At 1-month post-injury, animals will be sacrificed and tissue will be processed to evaluate inflammation and A $\beta$  deposition using proteomic and immunohistochemistry analyses as described in Aim 1.

Rationale for using 3xTg-AD model: This model contains APP<sub>Swe</sub> and tau<sub>P301L</sub> transgenes and PS1<sub>M146V</sub> knock-in mutations to mimic the key hallmark pathologies of AD including extracellular A $\beta$  plaques around 6 months, tau hyperphosphorylation around 6 months, and neurofibrillary tangles around 15months [17]. Cognitive deficits are first apparent around 6 months of age [17]. As such, a TBI at 10-12wks of age will offer a unique characterization of and how AD risk factors may interact with TBI sequelae contributing to acceleration of neurodegeneration.

Controlled Cortical Impact (CCI): The CCI model is a well-established rodent TBI model that imparts a moderate TBI [20,21]. Dr. Stabenfeldt's team is well versed in this model as demonstrated by recent publications [14,15]. Briefly, adult mice will be subjected to a unilateral contusion to the lateral frontoparietal cortex with the electromagnetic impactor. Trauma will be produced by activating an electromagnetic piston of 2 mm diameter 2 mm below the dura at 6.0m/s for 100ms duration. Sham groups will undergo the same surgical procedure for piston placement without the impact injury.

**Proposed One-Year and Long-Term Outcomes:** The results from this study will provide unique insight into the contribution of AD risk factors on BBB dysfunction and inflammation following TBI and vice versa (i.e., the contribution of TBI on AD pathology progression as relating to neuroinflammation and BBB stability). The collaboration between Drs. Stabenfeldt and Oddo, will ensure immediate access to the 3xTg-AD mice for the proposed study. Data and findings from this proposal will be disseminated at the appropriate national conferences and journal publications.

### **Year End Progress Summary:**

Aim 1: The Fall 2019 focused on completing all the proposed cohorts and tissue collection. We successfully completed these studies and are currently processing the tissue for cytokine profiling (in collaboration with Dr. Brafman), protein assessment (western blot), and also BBB permeability. We have validated the cytokine profile kit and antibodies for western blot assessment. Therefore, we are on track to complete or study by June 30, 2020.

Aim 2: For Aim 2, we modified the study design to compare the evolution of injury-induced neuroinflammation in the aged AD transgenic mice (20wks) to young adult mice (8-10wks). Therefore, we have begun the in vivo study for this cohort in February. The protein and histological assessment will mirror Aim 1 and thus we anticipate completing the study by June 30, 2020.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Project Report

**Developing A Univariate Neurodegeneration Imaging Biomarker with Morphometric Gaussian Process.** Yalin Wang, PhD, Richard J. Caselli, MD. Arizona State University; Mayo Clinic Arizona; Arizona Alzheimer's Consortium.

### **Project Description:**

Owing to the close relationship between neurodegeneration and cognition, atrophy measured by structural magnetic resonance imaging (sMRI) has been shown to quantitatively detect and track characteristic hippocampal, regional gray matter, and whole brain atrophy in clinical and late preclinical stages of Alzheimer's disease (AD). Currently, a single valued MRI atrophy measure is used as a neurodegeneration marker in the recently proposed AD descriptive "A/T/N" (amyloid, tau, neurodegeneration) system to define AD. However, the available *N* measures performed the worse among the three measures along the AD continuum [1]. Our current project's **objective** is to build and deliver a novel and highly sensitive univariate brain sMRI neurodegeneration index system that is clinically useful and able to expedite AD drug development by reducing clinical trial costs. **Hypothesis:** we hypothesize that this technique will better facilitate the identification of AD induced dementia and empower AD enrichment than previously shown by available competing algorithms, such as hippocampal volume, AD signature, and structural abnormality index (STAND)-scores, thus providing new imaging biomarkers as important outcome measures in clinical trials or for enrichment of individuals expected to progress within a particular time frame.

**Specific Aims:** To develop a robust and effective univariate neurodegeneration index and apply it to MRI brain scans of two well-characterized cohorts – the ADNI and AZ APOE cohort.

(a). Develop new geometry methods to compute morphometric Gaussian process-based index;  
(b). Validate the index by 1) correlation with AD genetic risk/AD severity; 2) progression prediction to AD or its prodromal stage via the Cox model; 3) sample size estimation for clinical trials.

### **Background and Significance:**

AD is the most common type of dementia [2]. It is generally agreed that effective presymptomatic diagnosis and treatment of AD could have enormous public health benefits. Neuroimaging research is currently focused on the development of accurate diagnostic markers that reflect the presymptomatic changes before the clinical onset of AD and can sensitively detect AD treatment effects in a sufficiently rapid and rigorous way [3-5].

Missing at this time is a widely available, highly objective univariate neurodegeneration biomarker capable of quantifying abnormal degrees of cerebral atrophy and reducing drug development costs in clinical trials. With much success in group difference study [6-8], including our own recent work [9-11], eventually one would like a method sensitive enough to apply to individual patients, compared against a normative sample. Therefore, a personalized brain morphometry measure based on an individual patient's brain scans with high diagnostic accuracy would be highly desirable for clinical use [12] and required by regulatory agencies for randomized clinical trials (RCT) [13]. Researchers in Arizona Alzheimer Consortium (AAC) pioneered the apolipoprotein E (APOE) e4 effect research in preclinical population [13-20]. We made the first strides into developing volumetric spectrum analysis methods [21-24]. Together we are uniquely positioned to develop a univariate neurodegeneration index system to quantify brain MR images for clinical research. The AAC grant, once available, will be leveraged to produce more exciting preliminary results. Our proposed project will make the planned R01 proposal submission more competitive.

## **Year End Progress Summary:**

*Computing Univariate Neurodegenerative Biomarkers with Volumetric Optimal Transportation.* In collaboration with Drs. Reiman, Caselli, Goradia and Chen, we propose a variational framework to compute optimal transportation (OT) on brain structural MRI volumes and develop a univariate neuroimaging Wasserstein Index (WI) based on OT to quantify neurodegenerative alterations. Experimental results, on 314 subjects (140 A $\beta$ + AD and 174 A $\beta$ - normal controls) from the Alzheimer's Disease Neuroimaging Initiative (ADNI) baseline dataset, provide preliminary evidence that the proposed WI is correlated with a clinical cognitive measure (the Mini-Mental State Examination (MMSE) score), and it is able to identify group difference and achieve a good classification accuracy, outperforming two other popular univariate indices including hippocampal volume and entorhinal cortex thickness. The current pilot work suggests the application of WI as a potential univariate neurodegenerative biomarker. This work has been accepted in Neuroinformatics, 2020.

*Applying Surface-Based Hippocampal Morphometry to Study APOE-E4 Allele Dose Effects in Cognitively Unimpaired Subjects.* In collaboration with Drs. Reiman, Baxter, Caselli and Chen, we characterized the ability of our automated surface-based hippocampal morphometry algorithm to distinguish between these three levels of genetic risk for AD. We examined the APOE-e4 dose effect on cross-sectional hippocampal morphology analysis in an MRI dataset, from AZ APOE cohort, consisting of 117 cognitively unimpaired subjects aged between 50 to 85 years (mean=57.4, SD=6.3), including 36 heterozygotes (e3/e4), 37 homozygotes (e4/e4) and 44 non-carriers (e3/e3). Our experimental results demonstrated its superiority to a commonly used hippocampal volume measurement. This work has been published in NeuroImage: Clinical, 2019.

*Integrating Convolutional Neural Networks and Multi-task Dictionary Learning for Cognitive Decline Prediction with Longitudinal Images.* In collaboration with Dr. Caselli, we propose a novel multi-task learning framework based on convolutional neural network (CNN). We applied this novel deep model on MRI data of 837 subjects from the ADNI dataset to predict future MMSE/ADAS-Cog scales. We also compared the prediction performances with seven other similar methods, and found that our method achieved superior results. Our work may add new insights into data augmentation and multi-task deep model research and facilitate the adoption of deep models in neuroimaging research. This work is under major revision in Journal of Alzheimer's Disease.

*A Univariate Persistent Brain Network Feature Based on the Aggregated Cost of Cycles from the Nested Filtration Networks.* We defined a univariate index which enjoys the monotonically increasing property to discover the dissimilarities among brain networks of AD, Early Mild Cognitive Impairment (EMCI), Late Mild Cognitive Impairment (LMCI), and normal control subjects (NC). We applied our method to ADNI dataset of Diffusion Imaging Tractography (DTI) brain networks of 200 subjects. The results based on different and same number of modules created in synthetic dataset show outperformance with respect to previous methods. Our work demonstrated promising results on classification between AD versus NC and MCIs. This work has been accepted in IEEE International Symposium on Biomedical Imaging: From Nano to Macro (ISBI) 2020.

*Diffeomorphic Smoothing for Retinotopic Mapping.* Prior work has demonstrated that retinotopic mapping may provide a way to quantify neurodegeneration, such as AD. We study a fundamental problem in retinotopic mapping: the alignment of visual cortical regions. We follow the common practice of cortical surface registration, i.e. the diffeomorphic condition: cortical surfaces can be aligned by stretching or/and shrinking but without tearing the cortical surface up. Specifically, the

diffeomorphic condition is quantified by the Beltrami coefficient. Then we modeled the registration as an optimization procedure of energy function, consisted of features (e.g. cortical thickness, surface curvature, etc.) similarity, and regularization (include the smooth and diffeomorphic constraints). We provided numerical steps to solve the optimization problem. We tested our registration on a synthetic dataset and surfaces with retinotopic data from the Human Connectome Project. We compared our method with popular surface/image registration methods for the synthetic data, including TPS (mean error in visual coordinates 4.47, the similar meaning of these numbers for later methods), D-Demos (1.20), LDDMM (0.64), and QCHR (0.10). We found the proposed algorithm achieves the smallest registration error (0.08) and ensures the diffeomorphic condition. We also apply the method on the retinotopic data from the Human Connectome Project retinotopic dataset. We achieved a better result than Freesurfer. Also, based on our method, we further improved a template for the visual cortex. This work has been accepted in ISBI 2020.

*Deep Multimodal Brain Network Learning for Joint Analysis of Structural Morphometry and Functional Connectivity.* We designed a supervised deep model to jointly analyze brain morphometry and functional connectivity on the cortical surface and we name it deep multimodal brain network learning (DMBNL). Two graph based kernels, i.e., geometry-aware surface kernel (GSK) and topology-aware network kernel (TNK), are proposed for processing the cortical surface morphometry and brain functional network. The vertex features on the cortical surface from GSK is pooled and feed into TNK as its initial regional features. In the end, the graph-level feature is computed for each individual and thus can be applied for classification tasks. We test our model on a large autism imaging dataset. The experimental results prove the effectiveness of our model. This work has been accepted in ISBI 2020.

*A Deep Learning Model to Predict Drug-gene Interaction.* We developed a deep learning-based model which takes advantage of information fusion to exploit essential feature of drug-gene pairs for DTI prediction. Firstly, since drug data and gene data have different feature distributions, two PCAs are used to extract individual features in their latent spaces, and also reduce dimensionality. Then, based on the globality of Fully Connected networks (FNN) and locality of CNN, we employ the FNN and CNN blocks to learn individual features from PCA features at the same time. Following the separated feature extracting layers, there are two information fusion layers. The first information fusion layer is utilized to merge outputs of CNN and FNN blocks and another one is adopted to mapping drug and gene features into one unified latent space. We believe that the fusion of those features can be more conducive to distinguishing the interaction of drug-gene pairs. We believe the model would be applied to discover putative drugs that can target over 600 AD associated genes. We apply the proposed deep model into drug-gene interaction predictions. Experimental results demonstrate our model can be applied as a potential classifier on drug-target interaction tasks.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

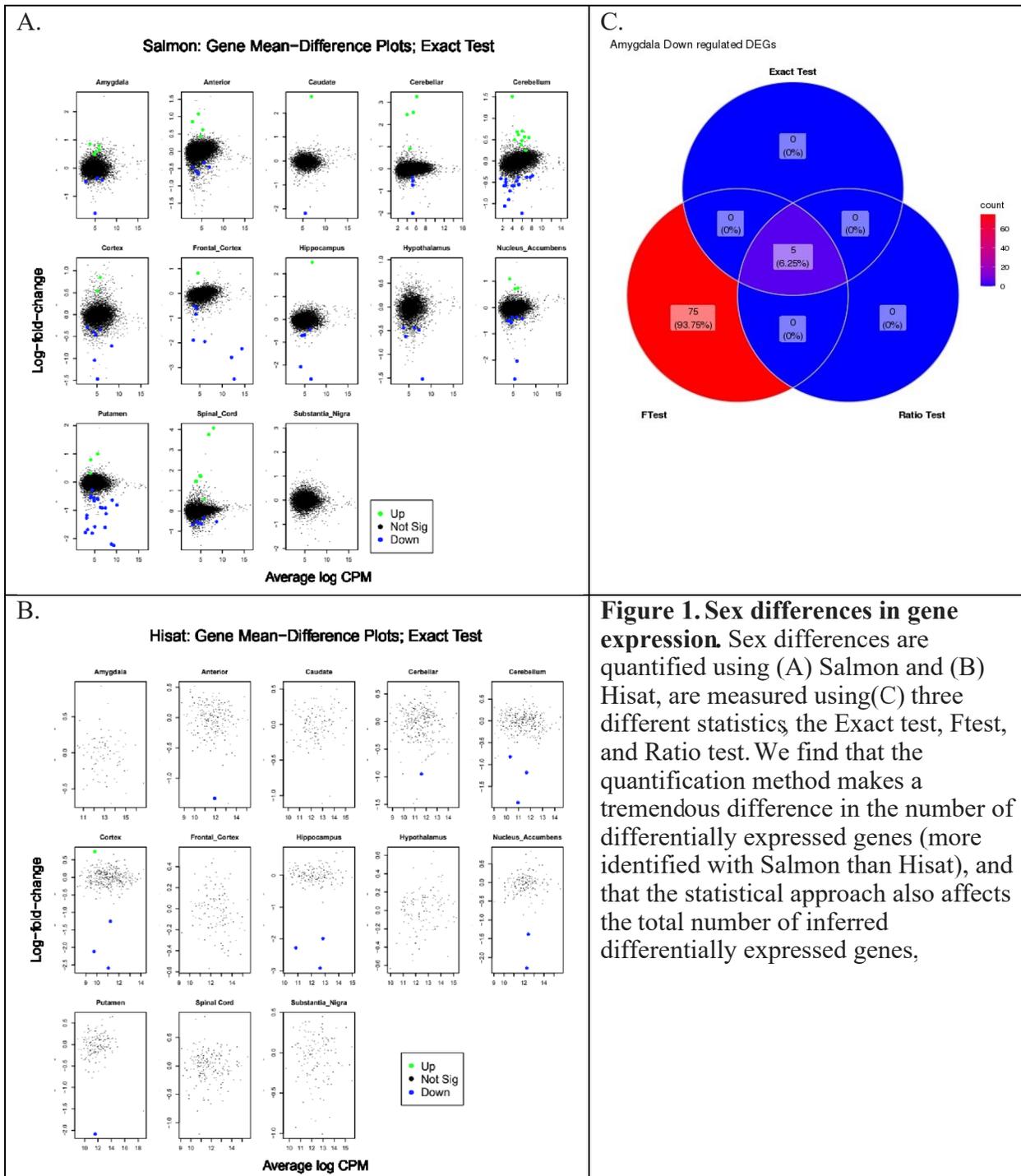
**Sex differences in healthy and Alzheimer's Disease brain gene expression.** Mollie Peters, MS, Heather Bimonte-Nelson, PhD, Salvatore Oddo, PhD, Melissa Wilson, PhD. Arizona State University; Arizona Alzheimer's Consortium.

### **Project Description:**

Alzheimer's disease is a form of a dementia that causes irreversible brain tissue damage that affects an estimated 5.4 million Americans (1). Currently, women at the age of 65 have a 1 in 6 chance of developing Alzheimer's disease while men of the same age have a 1 in 11 chance (2). Sex differences in gene expression in the brain are likely due to a combination of genetic (females have two X chromosomes while males have one X and one Y) and hormonal differences (gonadal hormones such as testosterone, progesterone and estrogen are notably different between the sexes). Notably, the most common risk allele for AD is the APOE-e4 allele, but this allele shows sex differences in penetrance and severity, increasing the risk of developing AD significantly more for female carriers than for male carriers, both in the heterozygous and homozygous states (3). Thus, the effect of the APOE4 allele is modified sex (4), resulting complicated sets of interactions affecting AD etiology. Genetic sex differences in gene content and expression have been shaped by millions of years of evolution. My lab has studied the evolution of these differences, and is now working to integrate this view with our understanding of human health. In particular, in this proposal we aim to study genetic and expression sex differences in healthy brains and brains from patients with Alzheimer's disease (AD) in an effort to pinpoint the mechanisms contributing to the sex differences in development and progression of AD. In our preliminary analysis we've found that there is substantial variation across brain regions in the magnitude of sex differences. In this proposal, we will narrow in on the specific patterns of sex differences across healthy brain regions, investigate sex-by-genotype interactions with the APOE locus, and validate our findings in two brain regions that we have biological replicates for. With our collaborators, we will be able to further functionally validate these interactions.

### **Year End Progress Summary:**

In the 2018-2019 year we developed a reproducible snakemake pipeline to process the more than 1,300 healthy brain samples from GTEx (Figure 1A); in 2019-2020, we extended this pipeline to include two sets of quantification software (one that is reference-based and one that is reference free), and began working with Dr. John Fryer to better focus on the sets of genes that are important for Alzheimer's Disease. We had previously downloaded and decrypted 3Tb of data for the brain samples from 13 different regions of the brain onto the ASU Research Computing cluster. All samples have been assessed for the library sizes and read lengths, and this year quality controlled all of that data, excluding less than 1% of samples due to poor quality (largely due to a long interval between time of death and RNA extraction). We find now that there are some tissues with significant differences in sex-biased gene expression (Figure 1), and others with almost no sex differences in gene expression. These results have been shared at the Genome Informatics 2019 meeting in November, and are currently being written up.



**Figure 1. Sex differences in gene expression.** Sex differences are quantified using (A) Salmon and (B) Hisat, are measured using (C) three different statistics, the Exact test, Ftest, and Ratio test. We find that the quantification method makes a tremendous difference in the number of differentially expressed genes (more identified with Salmon than Hisat), and that the statistical approach also affects the total number of inferred differentially expressed genes,

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Latino, Native American, and Other Research Participant Recruitment, Engagement and Retention.** David Coon, PhD, Richard Caselli, MD, Thomas Beach, MD, PhD, Lori Nisson, LCSW, Alireza Atri, MD, David Weidman, MD, Meredith Wicklund, MD, Geoffrey Ahern, MD, Steven Rapcsak, MD, Eric Reiman, MD. Banner Alzheimer's Institute; Mayo Clinic Arizona; Banner Sun Health Research Institute; Barrow Neurological Institute; University of Arizona; Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

This proposal will support the following specific aims.

- 1) Support new outreach strategies to increase enrollment and retention efforts
- 2) Provide subject reimbursements to increase enrollment and retention efforts.

### **Background and Significance:**

This proposal requests complementary support to enhance ongoing efforts for participant recruitment and outreach efforts as part of the Arizona Alzheimer's Research Consortium's ADC and ancillary programs. The Arizona ADC is part of a multi-institutional state-wide consortium that links together the major research institutions in Arizona to advance effort in the early detection, tracking of progression, and evaluation treatments and prevention therapies for Alzheimer's disease and related disorders. The ancillary programs include the Arizona BBDP and Arizona APOE4 Gene Dose Program. The Arizona Brain and Body Donation Program (BBDP) provides an invaluable scientific resource of longitudinal cognitive, motor, clinical, and genetic data from >800 living older adults who have standardized annual assessments, consent to brain (and frequently body) donation, and provide a resource of unusually high-quality brain tissue, postmortem CSF and blood samples (which differ in some respects to samples that are acquired in life) and neuropathological data after they die. The program includes but is not limited to research participants with the clinical features of Alzheimer's disease (AD) or related disorders and cognitively and neurologically unimpaired older adults with support from the National Institute on Aging (NIA)-supported Arizona AD Core Center (ADCC), research participants with the clinical features of Parkinson's disease (PD) and related disorders and cognitively and neurologically unimpaired older adults with support from the National Institute of Neurological Disorders (NINDS)-supported National Brain and Tissue Resource for PD and Related Disorders (NBTR-PD). The Arizona APOE4 Gene Dose Program provides an invaluable scientific resource of longitudinal data from initially cognitively unimpaired research participants with two, one and no copies of the APOE4 allele, the major genetic risk factor for AD. The program includes nearly 200 participants who were initially late-middle-aged participants with a first degree family history of dementia who are followed every two years with a battery of clinical ratings, cognitive tests, FDG, amyloid and now tau PET scans, and MRIs, who have provided plasma, serum and PBMC samples that are stored at Mayo Clinic, and who have begun to provide CSF samples with support from a longstanding NIA grant. It also includes more than 200 other participants, with or without a family history and through youngest to oldest adult ages, who are followed using state and organizational Arizona Alzheimer's Consortium funds, and who have not yet provided CSF, plasma and serum samples.

**Latino and Native American Enrollment:** Special attention has been given to minority enrollment to address the aims. The inclusion of participants with different characteristics will assist investigators in providing answers to questions about dementia diagnosis, treatment, and

management strategies that are likely to be applicable to the broad U.S. population. Additionally, a more diverse participant pool will facilitate investigations of different risk factors, health disparities and the neuropathology and genetics of AD and related dementias as well as studies of care giving and family burden in diverse groups.

**Proposed One-Year and Long-Term Outcomes:**

The proposed outcomes would be to increase enrollment and retention into the ADCC and its ancillary programs.

We will obtain IRB approval to provide subject reimbursements and begin providing reimbursements to eligible subjects.

Funds will be used in a way that complement but do not overlap with funding provided by the NIA and other funding sources for the ancillary programs. \$75 for everyone in the clinical core who agrees to a) amyloid PET, tau PET, MRI, blood, and/or b) brain donation. CSF is strongly encouraged when appropriate but not required for this compensation. \$30 to everyone else who completes UDS assessments. Currently this not funded by other funding sources. The other funding sources provide reimbursement for scans, blood and CSF collection that are supported by other funding sources.

**Year-End Progress Summary:**

Outreach efforts have included a combination of in-person events as well as traditional media and social media strategies. The in-person events range from tabling events at health fairs to community education presentations. Tabling events have been offered in faith-based organizations, AARP Latino events, Amigo Block Watch meetings, and at a variety of conferences. Community education presentations have been offered at HOPE Network meetings, in partnership with agencies such as the Area Agency on Aging, AARP, Banner Alzheimer's Institute, and multiple senior centers across the Valley. Social media outreach efforts included Facebook Live Events on multiple bilingual health/culture pages, bilingual newspapers/magazines, radio shows on Latino stations (i.e. AARP Latino station, La Reyna, and La Onda). Outreach on social media also heavily relies on cross-promotion from community partners; the most successful cross-promotion campaign was the partnership with Banner En Español, which helped us reach over 2,000 people through their post. Another recruitment effort that was utilized were postcards sent to areas, targeting people over the age of 50 who self-identify as Latino or Hispanic and whose primary language is English. In total, 5,000 postcards were sent to Latinos near the study sites. The HOPE Network and ASU team members have a list of 60 potential Latino/Hispanic participants referred to the ADC Aging study and they maintain contact with those individuals to help link them to study sites. Due to COVID-19, several participants have reported back that their study visits were postponed or cancelled until further notice; we will continue to stay connected with the interested AADC participants during the waiting period to ensure engagement once the study resumes. There have been ongoing discussions with Mindcrowd and All of Us about ways to target Latino/Hispanic and Native American participants to encourage enrollment in Arizona Alzheimer's Consortium activities. Due to COVID-19, we are investigating other platforms – such as video conferencing (i.e. Zoom), calling, emailing, texting, etc. – to provide outreach events and activities to reach these populations. The HOPE Network promotores have been able to successfully offer webinar series to the Latino community during the ongoing pandemic.

Outreach and recruitment funding also went to Mayo Clinic, Banner Health, Barrow Neurological Institute and the University of Arizona. Each of the sites was able to use these additional funds for outreach activities and to retain participants as well as increase recruitment and enrollment of

new participants, particularly Latino and Native American participants. Many of the sites worked in coordination with Dr. Coon's team. As with all projects involving human subject participants, the recruitment has slowed considerably due to COVID-19 and each team is reviewing next steps based on their site and its needs.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Website Update/Upgrade.** David W. Coon, PhD. Arizona State University; Arizona Alzheimer's Consortium.

### **Project Description:**

The project focuses on bolstering the Arizona Alzheimer's Consortium's (AAC) web presence through a comprehensive redesign and refresh of azalz.org.

### **Specific Aims:**

- 1) Increase accessibility by diverse populations, with key portions of a new "public engagement" sections available in both English and Spanish, and updated dynamic technology that meets accessibility standards (screen readers and more) for people with disabilities.
- 2) Ensure a clean and intuitive design that meets or exceeds the expectations of modern audiences, with additional media content (particularly video content) driving increased levels of public engagement.
- 3) Position azalz.org for healthy, organic future growth, maximizing search engine traffic and web visibility.

### **Designs and Methods:**

The project expands outreach efforts, taking the website from scientist-centric content to develop a new and media-rich public engagement section for lay audiences in diverse populations, with key content (including video content) developed in both Spanish and English. The effort does not duplicate material published on member institution sites, but instead creates complementary and supplementary material that answers the needs of our audiences, particularly as expressed and identified through surveys and Q&A data collected at the Consortium's public conferences. Finally, it maximizes methods for promoting AAC and member institution events and study opportunities, including standalone emails and targeted media content (videos, Q&As, interviews, documentary-style storytelling, etc.).

### **Year-End Progress Summary:**

The redesigned website is programmed and live on non-public "staging" server for review and testing, including dynamic functionalities for mobile platforms. There are more than 150 new images and graphics sourced for use as well as video content sourced and created to date that includes: BAI Native American Outreach; event coverage such as the HOPE Network *promotores* conference and interviews related to AD/DRD in the Latino community; motivational interviews with participants at the 2019 BAI Caregiver conference; informational interviews with physicians and community leaders filmed at the 2020 AAC Public Conference; ASU/Barrow Neurological Institute documentary-style memory loss storytelling and profiles of Latino caregivers; and, video content edited into 15 component videos for future use/publication to keep "public engagement" section dynamic. The project is on target for completion (publication to azalz.org by year's end) including Spanish language translation, assuming all necessary content is approved by May 2020. Longer term services include ongoing development of fresh, dynamic, bilingual material that will meet the informational needs of diverse and target populations, including underserved populations such as Latinos and Native Americans. Content will continue to be developed and refreshed in future years—making this not a one-off effort, but instead a substantial and ongoing push to expand public engagement, publicize research opportunities, and deepen institutional collaboration and connections.

**Project Progress Reports**  
**Banner Alzheimer's Institute**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Alzheimer's Prevention Registry.** Jessica B. Langbaum, PhD, Eric M. Reiman, MD, Pierre N. Tariot, MD. Banner Alzheimer's Institute; University of Arizona; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1. To increase enrollment into the Alzheimer's Prevention Registry through community outreach and other related efforts, particularly within Arizona.
2. To increase the number of study opportunities available to Alzheimer's Prevention Registry members, particularly within Arizona.
3. To compare the success rates of various approaches to promote study opportunities to Alzheimer's Prevention Registry members, tracking members' interest in each study opportunity.

### **Background and Significance:**

Enrollment and retention of participants are considered to be the biggest challenges researchers face. Current processes are generally inefficient, contributing to the expense and duration of trials. In the US, recent reviews show that 85-90% of all studies have delays in recruitment and enrollment, with 30% under-enrolling and only 7% of sites enrolling the projected number of participants in their originally stated timelines. Delayed or inefficient recruitment has scientific, financial, and ethical consequences. The web-based Alzheimer's Prevention Registry ([www.endALZnow.org](http://www.endALZnow.org)) ("APR") was created in 2012 to help studies meet their enrollment goals in an efficient and timely manner. At enrollment, individuals are asked to provide their email address and basic demographic information. Members receive regular email communication to keep them apprised of the latest news in Alzheimer's prevention research. In addition, members receive email notifications when study opportunities become available in their communities, with information on whom to contact to explore the possibility of their participation. In November 2015, the APR launched its GeneMatch program, an IRB approved research program open to adults age 55-75 in the United States who do not have a diagnosis of cognitive impairment. Upon enrollment into GeneMatch, participants are provided a cheek swab kit to provide a DNA sample for APOE genotyping, the results from which are used in part to help match to studies. As we continue to promote awareness of the APR and increase enrollment, it is imperative that we increase the number of and types of study opportunities available to members and compare the effectiveness of various approaches to promote study opportunities to members by tracking members' interest in each study opportunity. The results from this effort will provide invaluable information about the best way to raise awareness about study opportunities and recruit members for research studies.

### **Experimental Designs and Methods:**

To achieve Aim 1, we will work to expand Registry enrollment in Arizona. To achieve Aim 2, we will work with researchers across the country to request permission to add their studies to the Registry website, with a focus on studies led by Arizona Alzheimer's Consortium researchers. To achieve Aim 3, we will compare the effectiveness of various approaches for promoting study opportunities. We will track referral and enrollment numbers and time to fill sites' enrollment goals to assess the ability of the Registry to accelerate enrollment.

### **Proposed One-Year and Long-Term Outcomes:**

Results from the Registry will be submitted for publication in peer-reviewed journals and presented at scientific meetings. We will continue to seek additional external, non-state funding from NIH, industry and philanthropic organizations to support our efforts.

### **Year End Progress Summary:**

The Alzheimer's Prevention Registry is an online community of individuals age 18 and older who agree to receive emails with information about Alzheimer's prevention related research updates as well as notifications about study opportunities within their communities. As of January 2020, the Registry had over 348,000 enrollees and GeneMatch(1) enrolled over 90,000. Enrollment in GeneMatch was paused in April 2019 and will reopen in mid-2020 once the new genetic testing lab is onboarded. A manuscript describing the design, rationale, and initial results from the APR is under review.

**Aim 1).** During the funding period, considerable effort was undertaken to increase enrollment into the Alzheimer's Prevention Registry (APR) through community outreach, partnerships, and other related efforts. As of February 2020, the APR is comprised of 25,511 Arizona residents. Since July 2019, 3,656 individuals have joined the APR, of whom 251 (6.9%) are Arizona residents. As part of our engagement strategy, APR members opt in to receive our monthly e-newsletter with the latest news and information in AD research. Of the >348,000 members, 86,175 are considered "actively engaged." In 2019, 12 monthly e-newsletters were sent to APR members. The average e-newsletter open rate was 45% (compared to nonprofit healthcare industry average of 16%); the average e-newsletter click rate (percentage of APR members who clicked on the email in relation to those who opened/viewed the email) was 24% (compared to the industry average of 1.6%). To date, approximately 54,000 members have been added to the re-engagement campaign, and nearly 10,000 (18.5%) have been successfully re-engaged. We received an R01 grant from the NIA in September 2019 entitled "Establishing the science behind Alzheimer's recruitment registries: opportunities for increasing diversity and accelerating enrollment into trials", data from which will help increase enrollment of men and individuals from underrepresented populations into the APR (and other AD-focused recruitment registries).

**Aim 2).** The APR has helped recruit for more than 82 AD-focused studies since its inception. Currently, we are assisting with 11 in person studies in Arizona, with 5 more in the startup process.

**Aim 3).** We continue to examine success rates of various approaches to promote study opportunities to Alzheimer's Prevention Registry members, tracking members' interest in each study opportunity. In June 2019, the "Find a Study" section was updated to include a "contact form" for studies. Under the contact form model, individuals visiting the APR website and who are interested in a study are asked to fill out the form with their name, email address and phone number, review and acknowledge the study's eligibility criteria and authorization for APR to share their contact information with the enrolling study team. Individuals do not need to have joined the APR in order to search for studies on the website or to complete the contact form. We provide each enrolling study with a dashboard to track their referrals (i.e., inquiries from people who submitted their information via the contact form). Under this contact form model, studies and/or sponsors are required to execute a Data Use Agreement (DUA) due to the transfer of Personally Identifiable information (PII). Implementation of this contact form model will allow us to collect the data necessary to compare the success rates of various approaches to promote study opportunities to APR members, tracking members' interest in each study opportunity.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Alzheimer's Prevention Initiative (API).** Eric M. Reiman, MD, Pierre N. Tariot, MD, Jessica B. Langbaum, PhD. Banner Alzheimer's Institute; University of Arizona; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1. To continue to conduct a preclinical Alzheimer's disease (AD) trial/surrogate marker development program in cognitively unimpaired autosomal dominant (ADAD) mutation carriers within 15 years of their estimated age at clinical onset (i.e., the API ADAD Colombia Trial).
2. To conduct a preclinical AD trial/surrogate marker development program in cognitively unimpaired 60-75 year-old APOE4 homozygotes (i.e., API Generation Study 1).
3. To conduct a preclinical trial/surrogate marker development program in additional cognitively unimpaired 60-75 year-old APOE4 homozygotes and amyloid- $\beta$  ( $A\beta$ )-positive APOE4 heterozygotes (i.e., API Generation Study 2).
4. To plan and secure funding for other preclinical treatment trials programs/surrogate marker development programs in cognitively unimpaired individuals who are at risk for ADAD or LOAD (e.g., our proposed API-A4 aducanumab prevention trial).
5. To continue to support registries designed to assist with participant recruitment.

### **Background and Significance:**

The Alzheimer's Prevention Initiative (API) was established launch a new era in AD prevention research, accelerate the evaluation of promising AD prevention therapies, and find and support the approval of effective prevention therapies in at risk person as soon as possible. It currently includes 1) the API ADAD Colombia Trial of the primarily oligomeric  $A\beta$  antibody therapy crenezumab in cognitively unimpaired PSEN1 E280A mutation carriers from the world's largest ADAD (NCT01998841), 2) API Generation Study 1 of the relatively selective BACE1 inhibitor CNP520 and the active  $A\beta$  immunotherapy CAD106 in cognitively unimpaired APOE  $\epsilon$ 4 homozygotes (NCT02565511); 3) API Generation Study 2 of CNP520 in APOE  $\epsilon$ 4 homozygotes and  $A\beta$ + heterozygotes (NCT03131453); and 4) our proposed API-A4 aducanumab prevention trial in cognitively unimpaired  $A\beta$ + adults. It also includes the Alzheimer's Prevention Registry to help inform stakeholders and support their enrollment in these and other prevention trials, GeneMatch to help identify and support the enrollment of APOE4 homozygotes, heterozygotes and non-carriers in prevention trials, programs to disclose and assess the impact of a person's genetic or biomarker risk, future prevention trials (TBD), and other efforts to find and support the approval and availability of AD prevention therapies. Non-overlapping state and institutional funds are used to support these and related efforts, complement our NIH, philanthropic, and industry support, and help to find and support the approval of a prevention therapy as soon as possible.

### **Experimental Designs and Methods:**

To accomplish these overall goals and Aim 1, we will continue to follow participant randomized into the API ADAD trial until the last participant enrolled completes 5 years of blinded treatment (a "common close" design) as well as begin collecting tau PET. To accomplish Aims 2 and 3, we will continue to enroll participants into the API Generation Studies 1 and 2. To accomplish Aim 4, we will continue to work with our A4 colleagues to discuss plans for a prevention trial. To accomplish Aim 5, we will continue to expand the API Colombia Registry, for PSEN1 E280A kindred members, the web-based Alzheimer's Prevention Registry, and the GeneMatch program.

### **Proposed One-Year and Long-Term Outcomes:**

Data and findings from this proposed project will be submitted for presentation at relevant scientific conferences and in peer-reviewed manuscripts. In addition, the results will be used to inform future API preclinical treatment trials. The API will continue to seek additional external, non-state funding from NIH, industry and philanthropic organizations to support our efforts to conduct trials in at-risk populations.

### **Year End Progress Summary:**

**1. API ADAD Trial.** With primary support from initial and competitive NIA renewal grants, philanthropy, Genentech and its parent organization Roche, the API ADAD Colombia Trial continued to meet its stated goals. 365 participants were screened, 252 participants (including 162 PSEN1 E280A mutation carriers) were enrolled, with the last participant enrolled in early 2017 (1-3). Retention has been extremely high, and the placebo-controlled trial is intended to continue until early 2022, when the last person has been treated for 60 months. In addition to other clinical, cognitive and biomarker assessments, we began to acquire mid-treatment tau PET scans during this funding year (follow-up tau PET scans will be acquired at the end of the study). In accordance with Collaboration for Alzheimer's Prevention (CAP) principles we and industry partners have developed mechanisms to share baseline trial data and analyses with the field in ways that protect research participant anonymity, confidentiality and genetic risk disclosure and clinical trial integrity, and we have an agreement to provide a public resource of trial data and biological samples after the trial is over. We shared baseline information from age-matched mutation carriers and non-carriers at the 2018 Alzheimer's Association International Conference (AAIC). Approximately 50% of the enrolled carriers had a negative A $\beta$  PET scan at the time of their enrollment, suggesting that trial participants may provide a particularly good test of the amyloid hypothesis, and we continue to analyze these and other cognitive, brain imaging and biomarker data from members of this kindred. To support the trial and related research studies, we and our Colombian colleagues continue to expand and leverage data and biological samples from the API Colombia Registry. During the past year, the Registry was used to support a range of studies, including studies showing a) the promise of plasma neurofilament light (NfL) measurements in the early detection and tracking of neuronal injury and/or neurodegeneration (Reiman AAIC 2019; Quiroz (submitted), b) the promise of plasma p-tau217 and p-tau181 measurements in the early detection and tracking of AD, (Palmqvist, submitted); and c) the demonstration of an association between two copies of the rare APOE3 Christchurch variant in a PSEN1 E280A mutation carrier who was resistant to the clinical onset of ADAD and the performance of related studies to suggest the mechanism by which APOE, its variants and future APOE-related treatments might exert their therapeutic effects(4). Several other manuscripts are in various stages of preparation.

**2. API Generation Study 1 and Generation Study 2.** The Alzheimer Prevention Initiative (API) Generation Program evaluated the effectiveness of the BACE1 inhibitor, umibecestat, and the active immunotherapy, CAD106, in delaying the onset of AD symptoms in APOE4 carriers(5). The Generation Program included two studies implemented in 23 countries at 207 sites. Recruitment in the program and treatment with umibecestat was terminated in July 2019 after detecting an early signal of mild worsening in some measures of cognitive function with umibecestat. At the time of discontinuation, 9623 participants have been recruited, more than 2700 completed the 12-week screening phase with amyloid (PET or CSF) testing; approximately 27% were APOE4 homozygotes (HM). About 60% of HMs and 35% of heterozygotes (HTs) had elevated brain amyloid. A total of 1623 participants were randomized: 478 to Generation Study 1 (all HMs) and 1145 to Generation Study 2 (including 20% HMs and 80% HTs with elevated amyloid). The last participant last visit is scheduled to occur in Q2 2020 after which the database will be locked. Data and samples will be analyzed and shared with the scientific community soon thereafter to help inform the design of future trials.

3. API A4 Alzheimer's Prevention Trial. In 2018, API and A4 leaders received a \$33M NIA grant to help support a proposed prevention trial of an A $\beta$ -plaque antibody in cognitively unimpaired A $\beta$ + adults. Although we originally intended to partner with Biogen and use aducanumab, in March 2019, Biogen announced the discontinuation of their two Phase 3 trials (ENGAGE and EMERGE) of aducanumab in patients with MCI due to AD and mild AD dementia. The decision to stop the trials was based on results of a futility analysis indicating the trials were unlikely to meet their primary endpoint upon completion. Soon thereafter Biogen decided to pause any further decisions on a preclinical/prevention trial of aducanumab. As a result, the API and A4 leaders entered discussions with another industry partner and are planning for a prevention trial of a different A $\beta$ -plaque antibody in cognitively unimpaired A $\beta$ + adults.

4. Participant Recruitment Registries. We continue to expand the Alzheimer's Prevention Registry (APR), a web-based registry focused on encouraging enrollment into prevention studies. The Registry has >345,000 enrollees and is actively recruiting for dozens of studies nationally. A manuscript is under review describing the design, rationale, and initial results from the APR. In November 2015, the Registry launched its GeneMatch program which collects genetic samples from participants age 55-75 for APOE genotyping and uses the genetic results in part to help match people to research studies(6). More than 90,000 have joined GeneMatch to date, though enrollment is paused while we onboard a new genetic testing lab. GeneMatch is also helping with recruitment for several studies. We exceeded our ambitious goals for the Colombian API Registry, to date having enrolled nearly 6,000 members of the Colombian PSEN E280A kindred, including nearly 1,200 mutation carriers (7), and we continue to advance the study of AD in members of this extraordinary kindred. We received 2 NIH grants (R01 and SBIR) in 2019 to support efforts to study the science of recruitment and understand motivators and barriers to enrolling men and individuals from underrepresented populations to APR and GeneMatch. In addition, we submitted a R33 grant in February 2020 entitled "*Optimizing Research Infrastructure of Registries to Accelerate Participant Recruitment into Alzheimer's Focused Studies.*"

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Advanced Image and Statistical Data Analysis Techniques for Detection and Monitoring of Alzheimer's and Related Disease.** Yi Su, PhD, Dhruvan Goradia, PhD, Hillary Protas, PhD, Michael Malek-Ahmadi, PhD, Yinghua Chen, MS, Wendy Lee, MS, Teresa Wu, PhD, Jing Li, PhD, Rong Pan, PhD, Kewei Chen, PhD, Eric M. Reiman, MD. Banner Alzheimer's Institute; University of Arizona; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) To further develop and validate techniques that has the potential of improving sensitivity and statistical power of detecting Alzheimer's Disease (AD) related pathologies.
- 2) To develop and validate machine learning based techniques that can harmonize amyloid PET measurements obtained from different tracers and explore additional applications of machine learning and deep learning and AD image analysis.
- 3) To further develop and validate tau and amyloid PET analysis approaches using network and graph theory.
- 4) To further develop and maintain automated image processing and analysis pipelines to facilitate AD research and data sharing.
- 5) To apply advanced imaging and statistical methodologies on Arizona APOE cohort, ADNI, API-ADAD and other AD and aging study cohort to improve our understanding of AD and related dementia.

### **Background and Significance:**

Although the disease mechanisms are still not fully understood, AD is characterized neuropathologically by extracellular amyloid accumulations and intracellular tangles of hyperphosphorylated tau protein, both of which can now be measured in vivo using positron emission tomography (PET) imaging. In addition, other AD related changes of the brain such as reduction in glucose metabolism, inflammatory microglia activation, and synaptic density changes can also be measured and monitored in vivo using PET. Reliable quantifications of these pathological events using imaging data is critical to the better understanding of AD and the development of successful management strategies. However, several technical hurdles are hampering the derivation of reliable and accurate imaging biomarkers from the raw data. 1) PET has inherently low spatial resolutions due to limitations of imaging physics, and hence, measurements derived from PET data are inaccurate due to artifacts caused by the low resolution. 2) While a number of F18 based PET tracers were developed and approved by FDA for in vivo imaging of amyloid burden, each tracer has its own chemical properties that results in inconsistent assessment of amyloid burden. This inconsistency is especially problematic when quantitative measurement is needed. 3) Unlike amyloid PET which are relatively uniform and highly correlated among different brain regions, Tau deposition can be highly variable both spatially and among different individuals, results in additional challenges in quantifying overall Tau burden and tracking Tau accumulation. We aim to address these challenges in this year's methodological development efforts and making tools available to the local and external research communities.

### **Experimental Designs and Methods:**

**Aim 1.** We will further develop and validate the adaptive sampling based voxel-wise partial volume correction technique using ADNI data. We will optimize the adaptive sampling procedure

by testing different sampling density and sampling scheme, i.e. random sampling vs. systematic sampling, and examining the ability to improve sensitivity for different clinical/preclinical subgroups of ADNI participant.

**Aim 2.** We will implement a deep learning based framework that are capable of generating synthetic amyloid PET data based on structural MR image and amyloid PET data of a different tracer. We will train this deep learning model using a cohort of 100+ participants who have amyloid PET data acquired using both PiB and Florbetapir from the Open Access Series of Imaging Studies (OASIS) brain imaging database (<https://www.oasis-brains.org/>) series 3. Our primary model will be built to generate synthetic Florbetapir PET data from PiB and MR images, we will then test this model using data from the GAAIN database for Centiloid analysis (<http://www.gaain.org/centiloid-project>) which had 46 participants with both PiB and Florbetapir PET.

**Aim 3.** We will further develop and validate the graph theory-based methods using Tau PET data from ADNI and Arizona APOE cohort. We will correlate the novel Tau measurements with conventional region-based uptake measure, and examine the ability of this approach to differentiate participants at different preclinical and clinical stages of AD. We will also examine the ability of this measurement to track longitudinal changes of Tau burden.

**Aim 4.** We will further develop and maintain the PET Unified Pipeline and SPM based quantitative image analysis pipelines developed by CIAL and making them available to investigators within and outside of the Consortium.

**Aim 5.** We will apply the advanced imaging and statistical methodology that are developed to relevant AD and aging study cohort to better understand the disease.

#### **Proposed One-Year and Long-Term Outcomes:**

In the upcoming year, we anticipate generating results that further demonstrate feasibility of the proposed techniques in Aim 1-3 and build the foundation for future development and optimization of these techniques. We aim to publish these results in high-impact journals. In long term, we aim to strengthen our imaging and statistical expertise and build a cluster of advanced tools that facilitate the investigation of AD and related disease and the successful development of disease modifying treatments. We also intend to pursue extramural funding to allow expansion of our methodology development efforts.

#### **Year End Progress Summary:**

During the past year we continue to develop, test and apply advanced image analysis techniques for the early detection, tracking, and differential diagnosis of Alzheimer's disease (AD) and related dementia, the clarification of AD risk factors, and the evaluation of disease-modifying and prevention treatments.

We continue to develop and refine our proposed voxel-wise partial volume correction technique and evaluate its performance using the NIH APOE cohort and the ADNI cohort. Results demonstrated superior sensitivity and statistical power to detect longitudinal changes in amyloid burden (presented in AAIC 2019). Additional analysis is ongoing to further evaluate the advanced partial volume correction technique in combination with image analysis pipelines specifically designed for analyzing serial imaging data (manuscript in preparation).

We also continue to work on the application of machine learning and deep learning techniques in AD imaging research. Using a transfer learning (TL) model capable of handling missing data we examined the accuracy of using this model to identify ADNI MCI subjects who were diagnosed as MCI due to AD (N=97) from the rest of MCI patients (N=107) based on multi-model imaging data including T1-weighted MR, FDG and amyloid PET. The TL based model achieved a sensitivity of 0.90 and specificity of 0.84, while a conventional logistic regression model had a sensitivity of 0.83 and a specificity of 0.83. Based on the same cohort, we further examined the ability of the TL model in identifying MCI participants (N=29) who were at risk for

converting to AD and achieved a sensitivity of 0.86 and specificity of 0.94, while a logistic regression model achieved 0.43 in sensitivity and 0.95 in specificity (reported in AAIC 2019). Using a T1-weighted MR dataset of 1029 (926 training and 103 testing cases) participants with an age range of 18-86 years, we trained a 3D convolutional neural network (CNN) to estimate the chronological age based on MR imaging data and achieved a mean absolute error (MAE) of 5.48 years and a Pearson's correlation of 0.95. In comparison, Cole et al. achieved a MAE of 4.16 years and Pearson's correlation of 0.96 on a larger dataset of 2001 participants. In a separate collaboration with colleagues at the Beijing Normal University, a similar approach was used to predict chronological age based on MR imaging data of specific brain networks instead of the full MR image, and the best result was obtained using the prefrontal network trained on a cohort of 1454, (1303 training and 151 testing cases). The optimal performance was a MAE of 5.55 years (published in *Frontiers in Neurology* 2020). We further tested the hypothesis that the difference between predicted age from structural MR and chronological age can be used as a biomarker to detect pathological aging. An age-adjusted neural network (AD-NET) which combines the age prediction CNN with chronological age and then use both the structural MR image and the age information to predict whether an MCI patient will progress to AD within three years. The proposed AD-NET approach was able to achieve a sensitivity of 0.80 and a specificity of 0.73 using only structural MR image and age. This performance was comparable to other machine learning and/or deep learning approaches that use additional information such as PET and fluid biomarkers (manuscript under review). In addition, we are in the process of analyzing PIB-Florbetapir crossover data from the OASIS-3 cohort to examine the feasibility of using deep learning approach to harmonize amyloid PET imaging data derived from different PET tracers. We are planning to further expand our research on the application of machine learning/deep learning techniques and pursue extramural funding support in the near future.

Using a set of nodes defined based on AAL regions, brain networks were constructed from individual tau PET images from the ADNI cohort, and network metrics such as efficiency and strength were determined and assessed for their ability to differentiate clinical stages of AD and correlate with cognitive measurements. We found network measures of efficiency and strength to be significantly different among clinical AD groups. In addition, we found tau network efficiency measures strongly associate with memory performance measures even in the cognitively unimpaired group. Evaluation of the tau network measurements is also ongoing to determine their sensitivity to longitudinal changes in tau pathology and the potential to serve as surrogate biomarkers in clinical trials.

Our team also continued to develop and maintain image analysis tools developed by our lab. In the past year, we performed calibration of two amyloid PET imaging analysis pipelines routinely used in the lab following the recommended procedures from the Centiloid Working Group and determined the linear transform equations to convert our target-to-reference ratio measurements for amyloid PET to the recommended Centiloid Scale which can facilitate comparison of analysis results from different centers and groups. We applied these conversions to the amyloid PET (264 PiB scans and 117 Florbetapir scans) analysis results from the Arizona APOE cohort. We also continued to process imaging data in the APOE cohort and using standardized pipelines such as FreeSurfer, PUP, and SPM and generate standardized outputs to facilitate data sharing. We also used these imaging data obtained from the Arizona APOE cohort and the Colombia ADAD cohort to evaluate and validate cutting edge development of novel blood-based biomarkers with three manuscripts currently under review or in preparation, which reported sensitivity of 0.91 in detecting amyloid PET positive cases using recently developed plasma A $\beta$ 42/40 ratio a sensitivity of 0.94 in identifying neuropathologically confirmed AD cases using a plasma pTau measurements; and a significant differentiations between ADAD mutation carriers from noncarriers as early as 22 years before clinical symptom onset using plasma NfL measurements.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Native American Outreach, Recruitment, and Retention Program.** David Weidman, MD, Lori Nisson, LCSW, Richard Caselli, MD, Edward Zamrini, MD, Eric M. Reiman, MD, Pierre N. Tariot, MD, and David Coon, PhD. Banner Alzheimer's Institute; Mayo Clinic Arizona; Banner Sun Health Research Institute; University of Arizona; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1. To forge a close working relationship with members of our Native American Community in the awareness, care, and scientific understanding of Alzheimer's disease (AD) through educational and service-related outreach activities.
2. To support the participation of interested Native Americans in the ADCC clinical core and research studies of interest to them without detracting from our other outreach and partnership-development goals.
3. To continue to work with our Native American partners to identify and begin to prepare for one or more research studies that advance the understanding of AD and/or service to patients and families from this understudied, underserved population.

### **Background and Significance:**

Native Americans facing the problem of Alzheimer's disease (AD) constitute the most underserved and understudied population in the United States. Since 2006, we have established an outreach program to help address the educational and clinical needs of patients, families and health care professionals; developed culturally sensitive educational and service programs; and demonstrated to the Native American communities our strong interest in serving these needs, whether or not they participate in research studies. We have continued to attract a number of interested participants from the Urban Native American community to participate in the Arizona Alzheimer's Disease Core Center (ADCC) Clinical Core.

### **Preliminary Data and Plan:**

To date, 72 Native Americans have been followed through the ADCC and whose clinical findings are reported in a national database. As of January 2020, there are 26 active participants, 44 have withdrawn, and 2 died. Through January of 2020, there have been many participants who became lost to follow up and withdrawn from the program. Over the past year, over 3215 Native Americans have participated in education and outreach efforts led by our team. We continue building working relationships with numerous Arizona tribes and have also been successful with outreach efforts from tribes outside of Arizona including Oregon, New Mexico, Wyoming, Colorado, Utah, California, North Carolina, Alaska, Hawaii, Oklahoma, Nevada and Minnesota. Our team has been invited to present at the National Indian Council on Aging Biennial Conference held in September in Reno, NV. We hosted the 15th Annual Conference on AD in Native Americans in October at Camp Verde, AZ with over 90 professional participants in our preconference intensive and 200 community participants at our full-day training. Our outreach efforts led to 80 referrals to the ADCC. In addition, we culturally sensitive Native American and Dementia Tool Kit to educate a group of 50 tribal professionals in Phoenix with a comprehensive collection of training materials to better equip them to provide education and support to tribal colleagues and families caring for persons with dementia. Our team was invited and presented our work at the National Indian Council on Aging Biennial Conference in Temecula, CA in September 2018 and has been secured to present several topics at the next conference being held in Reno, Tahoe in August 2020. In

addition, Banner Alzheimer's Institute's Native American Outreach program and educational resources are being featured as a resource in the updated Administration on Aging Title VI Resource Manual.

We have initiated interactions with the Strong Heart Stroke Study (SHSS), BNI, and the University of Arizona-Banner All of Us (AoU) Program to help increase recruitment and retention of Native Americans into our ADCC Clinical Core and to support their participation in productive and impactful research studies. BAI has helped our BNI colleagues with the acquisition of follow-up MRIs from Native American participants at BNI. It has been providing image analysis training, resources, and K01 co-mentorship of Astrid Suchy-Dicey to support the analysis of baseline MRI data. It has begun to meet with Dr. Coon, AoU, BNI, and SHSS colleagues to help recruit, retain, and promote the study of 100 active Native Americans in the ADCC Clinical Core by September 2020.

Our advisory committee has begun planning the 16th Annual Conference on AD in Native Americans in October in the Gila River valley southern region of the state and we anticipate drawing more than 200 community participants. We will continue outreach efforts across Arizona and will continue to educate professional and family caregivers within our tribal communities. We have established consistent and solid working relationships with many urban and tribal communities to continue with these efforts. We will hold at least six public events to promote awareness in urban and reservation communities and offer another Native American and Alzheimer's disease tool kit training as part of our community outreach events this year. Our outstanding Native American outreach staff and other colleagues will continue to establish close working relationships with stakeholders from many tribes and nations to expand outreach, education and offer available resources for care and research.

**Proposed One-Year and Long-Term Outcomes:**

1. Continue outreach efforts to general Native American communities and education of health care providers for American Indians that will decrease the disparity related to diagnosis and treatment of AD and related disorders, in both reservation and urban dwelling Natives.
2. Help to recruit and retain Native Americans into the ADCC Core, such that we are following >60 actively enrolled NAs at BAI and a total of >100 enrolled NAs at all clinical core sites, capitalizing in part on emerging relationship with our colleagues from the SHSS and AoU programs.
3. Refine methods to reach more Native Americans from youth to elders to educate using the Native American Brain Health program.
4. Increase national engagement, knowledge, and collaboration amongst clinicians and researchers treating Native Americans using data gathered through the study. Leverage available data for educational purposes at the Annual Conference on Alzheimer's disease in Native Americans.

Funds will be used in a way that complement but do not overlap with funding provided by the National Institute on Aging (NIA, which supports some of our outreach and clinical core enrollment activities), the Ottens Foundation (which provides partial support for our Annual Conference), and funds from Freeport-McMoRan Native American Partnership Fund to support development of culturally sensitive NA programs.

**Year End Progress Summary:**

**Aim 1:** During the past year, our education and outreach programs reached 1117 professionals 2098 community participants from the Native American tribal communities across Arizona. Many topics focused on Brain Health and caregiver wellness, Alzheimer's disease education and

encouraging tribal communities to adopt Dementia Friends programming to help raise awareness and lower stigma. We hosted the 15th Annual Conference on AD in Native Americans in October at Camp Verde, AZ with over 90 professional participants in our preconference intensive and 200 community participants at our full-day training. Our outreach efforts led to 80 referrals to the ADCC. In addition, we utilized our culturally sensitive Native American and Dementia Tool Kit to educate a group of 50 tribal professionals in Phoenix with a comprehensive collection of training materials to better equip them to provide education and support to tribal colleagues and families caring for persons with dementia. Our team was invited and presented our work at the National Indian Council on Aging Biennial Conference in Temecula, CA in September 2018 and has been secured to present several topics at the next conference being held in Reno, Tahoe in August 2020. In addition, Banner Alzheimer's Institute's Native American Outreach program and educational resources are being featured as a resource in the updated Administration on Aging Title VI Resource Manual.

**Aim 2:** During the past year, we enrolled 6 new participants, completed 26 total assessments, and 34 participants were lost to follow-up. We have 18 additional participants in Quarter 1 of 2020 who are scheduled to enroll. To help minimize attrition, we will be working with the ADCC Education Core to find ways to optimize retention in our longitudinal research program. With our SHSS partnership we anticipate recruiting and retaining interested Native American participants at a greater frequency throughout the year. We have continued to reach participants at community events, and we have begun to explore new relationships with partnering organizations to help in the recruitment, retention, and productive study of Native American research participants.

**Aim 3:** BAI Native American Program has received funding from the Ottens Foundation, which provides partial support for our Annual Conference and funds from Freeport-McMoRan Native American Partnership Fund to support development and advancement of culturally sensitive Native American education and support programs an extended.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Advancing the Use of Lumbar Puncture in Alzheimer's Disease and Related Disorders Research.** Danielle Goldfarb, MD, Alireza Atri MD, PhD, Daniel Viramontes, BS, Michael Callan, BS, Gene Alexander, PhD, David Weidman, MD, Po Tsai, MD, Marina Reade, DNP, Carolyn Liebsack, BSN, Eric M. Reiman, MD. Banner Sun Health Research Institute; Banner Alzheimer's Institute; University of Arizona College of Medicine-Tucson; University of Arizona; Translational Genomics Research Institute; Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

**Aim 1.** Train 8-12 Arizona Alzheimer's Consortium clinicians on ultrasound-guided lumbar puncture (Us-LP) techniques to implement Us-LP into their research practices.

**Aim 2.** Preliminarily assess the feasibility and utility of Us-LP implementation in ADRD research studies by a pilot program that measures clinician confidence, performance ratings and adverse events rates in Us-LP before and after clinician training and pre- and post-procedure.

**Aim 3.** Develop print and video materials for clinicians and the public on the importance, utility, and safety of LPs in order to enhance the perceived value; mitigate fears and misconceptions; and increase the likelihood of participation and referral for LPs.

**Background and Significance:** At this time, due to the amount of ADRD-related information obtainable in CSF samples, cost, and availability, LP represents the most promising ADRD diagnostic approach to support biomarker development research and to accurately diagnose AD for inclusion in prevention and treatment trials. Advances in CSF and neuroimaging biomarkers support the detection of pathology along the AD ATN research continuum<sup>4</sup>. While neuroimaging biomarkers have evolved dramatically in recent years, high costs and lack of accessibility remain barriers to widespread use of amyloid and tau PET; and the latter is currently only available in research settings. CSF analysis provides a unique window into brain pathobiology and allows simultaneous testing for multiple biomarkers of disease and injury, including amyloid and tau species, alpha-synuclein, inflammation, and axonal and synaptic injury.

The preponderance of evidence supports the utility of LPs for diagnosis of numerous neurological conditions, and LPs as safe and well-tolerated with low AE rates when performed correctly.<sup>5</sup> Yet, various patient-, provider-, and procedure-related factors contribute to lower success rates and higher LP-related AEs. A cross-sectional survey of primary care physicians and specialists across Europe and North America<sup>1</sup> reported CSF AD biomarker testing ordered by 26% of respondents, with only 6% of primary care physicians reporting affirmatively. There was great geographic variation, with higher use in Europe and with only 15% of US physicians ordering CSF. The major challenge identified was the invasiveness of the procedure. Other reasons included patient concerns, limited capacity to test universally, and inconclusive results. The LP program aims to proactively address these factors, specifically considering our older population, through education of patients, families and dementia specialists and referring clinicians.

Ultrasound technology is not yet used routinely with LPs. Studies have shown variable benefit for improving LP success rates; however, most studies evaluated Us-LP in emergency department and pediatric settings. Ultrasound is indicated when bony landmarks are difficult to palpate and when congenital or chronic spine issues are present<sup>6</sup>. A recent review and meta-analysis<sup>3</sup> reported Us-LPs were associated with higher success rates, fewer traumatic LPs, shorter time to success, fewer needle passes, and lower pain scores. They concluded that ultrasound should be considered for all LPs, particularly in individuals with difficult anatomy.

Successful implementation of this innovative pilot program will set the stage for dissemination to other ADRD centers in AZ and the US. These educational efforts could substantially impact ADRD outreach and research by mitigating barriers related to knowledge and stigma associated with LPs, and improve willingness to participate, advocate and refer.

### **Experimental Designs and Methods:**

Aim 1: In the summer of 2019, 8-12 Arizona Alzheimer's Consortium clinicians interested in incorporating Us-LP into their research practices will complete a course led by expert faculty at the Banner Simulation Laboratory, Phoenix, AZ.

Aim 2: Implementation of a pilot program to assess pre- and post-Us-LP training outcomes on LPs performed at BAI and BSHRI ADRD-related research including observational cohorts, diagnostic and biomarker studies, and clinical trials. Participants will be consecutively enrolled in the proposed study whenever an LP is considered for diagnostic, research or clinical trial purposes. Patients refusing LP will also be provided the opportunity to enroll in this project to determine LP-related attitudes and acceptance rate. Participants with contraindications for LP, including anticoagulant treatment that cannot be interrupted and evidence increased intracerebral pressure, will be excluded. Institutional Review Board approval will be received before the study commences. Survey forms developed at the Clinical Neurochemistry Laboratory, Salgrenska University, Mohlndal, Sweden<sup>7</sup> will be adapted; patient and clinician characteristics and attitudes; details of the LP procedure; use of ultrasound; and pre- and post-training clinician confidence, LP performance, and LP-related AE's will be measured. Since ultrasound will not be used by clinicians until after training, questionnaires will also be administered at a small number of LPs (n~15) prior to Us-LP implementation allowing for both pre- and post-training (between LP method) comparisons (15 LPs pre-training, 60 LPs post-training), and pre- and post-LP (pre/post within LP method) comparisons.

Aim 3: We will develop print and video educational materials on the nature, value, safety and tolerability of LPs along with the availability of ultrasound technology. Targeted materials will be developed for patients/families, referring clinicians, and the community at large and will be incorporated in community outreach including lectures, newsletters, articles, and local and regional campaigns. Patient, family and referring provider surveys and focus groups will assess pre- and post-education attitudes about LP and referral rates.

**Proposed One-Year and Long-Term Outcomes:** Outcomes data (clinician confidence and attitudes regarding LP and Us-LP; patient, family and referring clinician attitudes and likelihood to participate or refer for LP; LP AE's; LP clinician and patient experiences) and findings from the proposed project will be analyzed and presented at scientific conferences and in peer-reviewed manuscripts. These pilot data will support applications for funding through NIH, industry and philanthropic organizations to enhance global efforts to increase capacity for ADRD fluid biomarker acquisition. A long-term goal of this program is to provide a foundation for a future training program for a LP proceduralist<sup>8</sup> that can be a mid-level practitioner or RN in order to vastly expand capacity to obtain CSF in an effective, efficient and scalable manner.

### **Year End Progress Summary:**

*Aim 1. Train 8-12 Arizona Alzheimer's Consortium clinicians on ultrasound-guided lumbar puncture (Us-LP) techniques to implement into their research practices.*

Very good progress was made on Aim 1 – six LP clinicians were trained on Us-LP techniques and implemented Us-LP into their research practices.

Two ultrasound-assisted lumbar puncture (Us-LP) courses convened and were led by Banner expert ultrasound faculty Dr. Teresa Wu (Associate Professor of Emergency Medicine, UA

College of Medicine Phoenix, Banner Simulation Curriculum Director) and Dr. Jason Grimsman (Assistant Professor of Emergency Medicine). Course descriptions are as follows: (1) Introduction to Ultrasound-Assisted Lumbar Puncture and Advanced Us-LP. The introductory course (Two hours on 8/22/2019) was completed by six dementia specialist clinicians (5 neurologists and one advanced practice nurse from two Banner sites-Banner Alzheimer’s Institute (BAI) and Banner Sun Health Research Institute (BSHRI)), during which time, Drs. Wu and Grimsman provided a didactic session following by simulation opportunities using the hand-held ultrasound equipment on two human volunteers and on simulation models. Needles were not inserted on human volunteers. For the advanced Us-LP course (3 hours on 11/21/2019), four of the six above clinicians complete the advanced training which demonstrated the use of an ultrasound cart system in conjunction with the hand-held tablet ultrasound devices on study participants undergoing lumbar puncture.

***Aim 2. Preliminarily assess the feasibility and utility of Us-LP implementation in ADRD research studies by a pilot program that measures clinician confidence, performance ratings and adverse events rates in Us-LP before and after clinician training and pre- and post-procedure.***

Excellent progress has been made on Aim 2 – 57 LP participants have been enrolled through February 2020; LP clinicians have completed Us-LP study questionnaires; and data on adverse events has been collected.

Study investigators reached an agreement with the company Philips to use their Lumify hand-held tablet ultrasound equipment for this AAC-funded study. Philips provided two tablets with two corresponding transducers (linear and curved). After IRB approval in August 2019, the study was initiated at BAI and BSHRI. All study participants who agreed to lumbar puncture for various observational and clinical trials at BAI and BSHRI were consented and enrolled sequentially (n=57). LP participants characteristics are listed in Table 1. For each LP completed, the LP clinician had the option to use ultrasound assistance. After the procedure, the LP clinician completed a post-procedure questionnaire about clinician background, use of ultrasound, and procedure performance. LP participants were contacted by the research coordinator within five days to identify adverse events. The research coordinator entered all questionnaire data into RedCap. The design (content and timing) of the questionnaires (clinician surveys) was modified from pre-/post-LP to post-LP based on input from LP clinicians, and consideration of Us-LP training, IRB approval and grant timelines. Data analysis is planned for Spring 2020 with the results available by the end of the grant period of June 30, 2020.

Table 1. LP Study Participant Characteristics

	Site 1	Site 2
Total Subject Count	39	18
<b>Gender</b>		
Male	14	5
Female	25	13
<b>Age</b>		
50-64	2	11
65-74	16	3
75-84	17	4
>=85	4	0
<b>Ethnicity</b>		
African American or Black	0	1
Caucasian	38	17
Asian American or Pacific Islander	1	0
Hispanic or Latino	0	0
Native American	0	0
<b>History of Headache</b>		
None or Rare	34	14
Mild	6	2
Chronic	0	1
<b>History of Other Chronic Pain</b>		
None or Rare	37	10
Mild	2	6
Chronic	1	1
<b>Prior LP</b>		
No	28	7
Yes	12	10
<b>Attitude to Undergo LP</b>		
Very concerned	2	2
Somewhat concerned	7	5
Neutral	16	4
Somewhat unconcerned	8	3
Very unconcerned	7	3

*Aim 3. Develop print and video materials for clinicians and the public on the importance, utility, and safety of LPs in order to enhance the perceived value; mitigate fears and misconceptions; and increase the likelihood of participation and referral for LPs.*

Good progress has been made on Aim 3 - both print and video educational materials are under development. Two video production companies provided quotes for a brief education video. We anticipate these materials will be completed in the summer of 2020.

**Long Term Goals:**

Preliminary results so far suggest that Us-LP is feasible to implement in an AD/ADRD clinical research setting. Data analysis on the utility of Us-LP will be conducted in Spring 2020. Results will be submitted for presentation at the Arizona Alzheimer's Consortium Conference (if late abstract accepted), the 2021 Alzheimer's Association International Conference, and the 2021 American Academy of Neurology Meeting. Upon study completion and based on our findings, we plan to apply for additional funding through NIH, industry and/or philanthropic organizations to develop enhanced efforts to increase the use of lumbar puncture and fluid biomarker acquisition in ADRD research. We continue to anticipate that this study will provide a foundation for a future training program for a LP proceduralist that can be a mid-level practitioner or RN in order to vastly expand capacity to obtain CSF in an effective, efficient and scalable manner.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Biomarkers in the Risk for Alzheimer's Disease.** Gene Alexander, PhD, Connie Boker, Thomas Beach, MD, PhD, Richard Caselli, MD, Yi Su, PhD, Eric Reiman, MD. University of Arizona; Banner Alzheimer's Institute; Banner Sun Health Research Institute; Mayo Clinic Arizona; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

This highly collaborative, multi-institutional project proposes to leverage the unique cohort of healthy older adults enrolled in the Brain Body Donation Program (BBDP) at Banner Sun Health Research Institute (BSHRI), as well as ongoing efforts for the enrollment of Arizona Alzheimer's Disease Core Center (ADCC) participants in our Native American and Hispanic/Latinx cohorts. We will address the following specific aims: 1) to make available for research neuroimaging scans from clinically well-characterized, cognitively unimpaired older adult cohorts; and 2) to develop and implement novel methods for processing and analysis of neuroimaging scans to identify new multimodal imaging biomarkers for cognitive decline and preclinical AD risk. Additionally, this study provides added value by providing an opportunity to help: 1) create a unique dataset to support cognitive aging and AD research across Arizona, 2) explore how brain imaging measures relate to demographic and cognitive differences in older adults; 3) evaluate how neuroimaging biomarkers ultimately relate to post-mortem brain pathology, and 4) support new external grant proposals on aging and AD risk.

### **Background and Significance:**

The population of older adults is expected to grow rapidly over the next two decades and with this growth of the aging population, it will be increasingly important to respond to the associated increases in AD, both in Arizona and nationally. Recent advances in PET imaging have allowed the pathological features of amyloid- $\beta$  (A $\beta$ ) plaques and tau neurofibrillary tangles to be detected in living humans. We now have extensive local experience in the Arizona ADC with radiochemistry methods needed to produce  $^{11}\text{C}$  PiB and several  $^{18}\text{F}$  labeled tracers for amyloid and tau imaging, providing valuable markers of AD pathology in individuals prior to the development of cognitive symptoms. This proposal leverages the expertise and infrastructure of the Arizona Alzheimer's Consortium and ADCC Brain Imaging and Fluid Biomarkers (BIFB) Core, as well as our ongoing longstanding studies, allowing us to efficiently combine brain imaging and fluid biomarkers of amyloid and tau with other biomarkers and cognitive measures. These measures will ultimately be of even greater value due to their potential linkage to neuropathology in many of our participants, providing an opportunity for deep phenotyping that exponentially exceeds the typical clinical diagnosis, allowing for a unique precision medicine approach to neurodegenerative disease. Given that we plan to include participants from our BBDP, we also expect to ultimately have the opportunity for neuropathology validation for imaging biomarkers in a subset of our proposed sample. Neuroimaging and fluid biomarkers will be integrated into our ongoing ADCC efforts with cognitively normal participants who are now approaching peak years for AD. This work will serve to further establish and expand the infrastructure for the BIFB Core across the Arizona ADCC, while providing critical data needed to inform analyses and statistical power for the use of biomarkers in prevention trials. Together, these data will also provide a valuable resource for developing and testing new fluid biomarker assays in relation to brain imaging and ultimately to neuropathology findings.

### **Experimental Designs and Methods:**

This project proposes to leverage the unique cohort of healthy older adults enrolled in the BBDP led by Dr. Tom Beach, as well as ongoing efforts for the enrollment of ADCC participants in our Native American and Hispanic/Latino cohorts. These participants already receive annual clinical and cognitive assessments and some have agreed to post-mortem donation of their brain to support aging and AD research, as part of the Arizona ADCC. Additionally, a portion of the BBDP cohort is already planned for recruitment to participate as part of the ADCC BI-FB Core. The BIFB Core includes several collaborators from this Arizona Alzheimer's Consortium (AAC) proposal (Alexander, Beach, Reiman, Su). We expect to collect amyloid and tau PET and MRI scans in approximately 27 cognitively unimpaired individuals drawn from the BBDP, as well as from the Native American and Hispanic/Latino Cohorts. Participants will be contacted during their annual clinical/cognitive assessment to schedule amyloid PET and MRI scans. Data collected from these participants will be uploaded to the ADCC Database for quality control procedures and archiving and will be linked to their clinical and cognitive data obtained during their annual visit.

### **Proposed One-Year and Long-Term Outcomes:**

This work will be leveraged to support complementary projects investigating neuroimaging effects of preclinical AD and how these effects are influenced by difference in demographic diversity. These studies reflect collaborations focused on developing externally funded grant proposals, as part of a multi-disciplinary, collaborative research program that will leverage the expertise of investigators across the Arizona ADCC. The proposed research will provide novel and rich datasets leading to published findings to advance our understanding of brain changes associated with the risk for dementia and cognitive decline. Importantly, it is expected that this dataset will provide essential pilot data to support new proposals for external NIH funding. Specifically, this project will provide key data and methodological developments to help support our planned competitive NIA ADCC renewal submission, as well as other grant applications that utilize brain imaging biomarkers of preclinical AD.

### **Year End Progress Summary:**

We have made significant progress in our research efforts supported by this project to advance the availability, use, development, and implementation of biomarkers to assist Alzheimer's disease research across the state of Arizona. During this past year, this project has provided critical support needed to augment and enhance efforts for the collection of neuroimaging scan data, including positron emission tomography (PET) with amyloid (PIB and florbetapir) and tau (flortaucipir) tracers, as well as magnetic resonance imaging (MRI) of brain structure, function, and connectivity, in participants from the BSHRI BBDP. This project has also additionally helped to augment our efforts to collect blood and cerebrospinal fluid (CSF) samples for storage and testing of key fluid biomarkers in this novel cohort of older adults who have agreed to donate their brain and body for research after death. This AAC effort, as part of ongoing plans for the ADCC BIFB Core, will provide a unique cohort of cognitively-unimpaired older adults to evaluate the relation of brain imaging and fluid biomarkers with brain pathology obtained after autopsy. In addition, this AAC project has also helped to enhance and support our ongoing efforts to increase diversity with greater inclusion of participants from the Arizona Native American and Hispanic/Latinx communities. Through efforts at BAI, increased outreach during the past year has implemented and helped strengthen a relationship with the Strong Heart Study, providing the opportunity to enroll Native American participants from the Phoenix community into our ADCC Clinical Core and BIFB Core. Similar outreach efforts in the Phoenix-metro area have also been implemented to enhance enrollment of Latinx participants for enrollment in both the ADCC Clinical Core and BIFB Core. Data collected from these participants are uploaded to the ADCC Database for quality control assessment and archiving, and will be linked to their clinical and cognitive data obtained during their Clinical Core visits. Together, we expect that these important

complementary efforts, supported by this AAC state project, will provide an important added contribution for the cohort collected here in Arizona, with benefits for the state of Arizona and across the Alzheimer's Centers nationally. Importantly, this effort will also greatly enhance and support our plans for our center grant application for the upcoming competitive renewal of the NIA ADCC proposal in fall of 2020.

This AAC project has also been part of the ongoing, integrated efforts to support the development, testing, and implementation of additional and novel brain imaging and other biomarker methods that may further advance and support collaborative research efforts both here in Arizona and nationally. Efforts at BAI, in collaboration with the ADCC BIFB, have focused on implementing a new and more cost-effective PET amyloid tracer (florbetaben), which has the potential to greatly enhance the availability and use of amyloid PET imaging for research investigators in both the Phoenix and Tucson-metro areas. It is expected that this new PET amyloid tracer will be available for implementation in both Phoenix and Tucson this year and will be included in the battery of neuroimaging scan options, as part of the ADCC BIFB Core. It is also expected that this methodological advance will further support our plans for the submission of the upcoming NIA ADCC competitive renewal application in fall of 2020.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Enhancements to a Centralized Data Management System for the Arizona Alzheimer's Disease Core Center (ADCC), Brain and Body Donation Program (BBDP), and Apolipoprotein E4 (APOE4) Gene Dose Program.** Don Saner, MS, Ricardo Amador, MS, Matthew Huentelman, PhD, Bruce Petersen, BS, Thomas Beach, MD, PhD, Richard J. Caselli MD, Eric M. Reiman, MD, David Coon, PhD, Dave Parizek, BS and colleagues from each of the participating sites: Banner Alzheimer's Institute; University of Arizona; Arizona State University; Mayo Clinic Arizona; Banner Sun Health Research Institute; Translational Genomics Research Institute; Barrow Neurological Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1) Enhance our infrastructure for Data, Imaging and Sample sharing which will include making our online Data Discovery tool available to the research community. The tool has a simple adhoc query mechanism for investigators to get counts of participants with Sample and Image Availability as well as the ability to submit and track requests.

2) Include data from our affiliated core projects, APOE Gene Dosing and Brain and Body Donation Cohorts on existing data sharing platforms, GAAIN and LONI, to enhance the visibility of our programs and provide another option for Data, Sample and Image sharing.

3) Complete the upload of 25 years of imaging from our programs to our Imaging Analytics platform, XNAT.

4) Support the new Bioimaging and Fluid Biomarkers core and other similar initiatives by creating REDCap forms to track participant information, Imaging meta-data and samples and incorporate this data into our centralized database

### **Background and Significance:**

The Arizona Alzheimer's Consortium has three longitudinal research programs which are internationally recognized for their productivity, impact, and value to researchers inside and outside of Arizona in the scientific fight against AD, PD, and related disorders, and the study of normal brain aging. These programs include common data elements, are administered through separate data management programs, and could provide even greater value under a common data management program that is optimized to fulfill the programs' common and complementary research goals. a) With support from the National Institute on Aging (NIA), the Arizona ADCC Clinical Core is the nation's first NIA-sponsored AD Center with multiple clinical core sites (including those at Mayo Clinic Arizona, BSHRI, UA, BNI, and BAI); it provides annual assessments in ~500 research participants with AD, related disorders, and cognitively unimpaired older adults; it includes individuals who are enrolled in BSHRI's BBDP, cognitively unimpaired individuals with two, one and no copies of the APOE4 allele, the major genetic risk factor for AD, members from Arizona's understudied Latino and American Indian communities, and other clinically affected and unaffected research participants; and it provides a shared resource of participants and data for researchers to generate new findings, publications, and grants. b) The BBDP includes >800 annually assessed research participants from the ADCC, the National Institute for Neurological Disorders (NINDS)-supported National Brain and Tissue Resource for PD (NBTR-PD), and other longitudinally assessments from older adults who consent to brain donation after they die, neuropathological data and exceptionally high quality brain and body tissues from >1,500 expired BBDP participants; it has been the world's leading resource of neuropathology data and brain and other body tissue samples for AD, PD and other neurodegenerative disease researchers around the world, has contributed to hundreds of

research publications and grants, and continues to make major contributions to the study of AD, PD, related disorders, and brain aging. c) With support from NIA, the state of Arizona and Mayo Clinic and BAI, the Arizona APOE4 Gene Dose Program provides a longitudinal cohort of research participants and data with two, one and no copies of the APOE4 gene, reflecting three levels of genetic risk for AD, including a sub-set of subjects with extensive brain imaging and other biomarker data. This program has made pioneering contributions to the conceptualization of “preclinical AD,” established a foundation for the Alzheimer’s Prevention Initiative (API) and the accelerated evaluation of prevention therapies, and includes an invaluable resource of data and samples to help researchers detect and track the earliest biomarker and cognitive changes associated with AD, contribute to the understanding of genetic and non-genetic risk factors, develop data analysis techniques with improved power to detect and track AD and evaluate promising but unproven AD prevention therapies. Consortium researchers lead other valuable longitudinal research programs, which despite fewer common data elements, may benefit from either a shared data management program and/or mechanisms to find other relevant data in the future. In this project, we propose to enhance the work done in the previous year on a centralized robust data management platform to include more real time reporting, include more data sources, optimize the code that extracts data for NACC submissions to include data consistency checks and create a data sharing platform.

#### **Experimental Designs and Methods:**

In the coming year we will continue to build out infrastructure to support sharing of Data, Biospecimens and Images with the research community. Key to this effort will be completing our Data Discovery tool which enables researchers to create ad-hoc queries against our centralized database to return counts of participants matching search criteria with Biospecimen and Image availability. Currently, the application lacks a robust request, tracking and fulfillment system which we will implement prior to the roll out to the research community. In addition to our Data Discovery tool, we are also working towards sharing our data from our ancillary cores with USC’s LONI and GAAIN platforms. This will give our programs greater visibility and will also permit us to link back to our Data Discovery tool. We have started the work of creating a data dictionary and mapping our data to their required format. In parallel, we are working with legal/regulatory to ensure proper consents and legal agreements are in place to permit sharing. As part of our Data, Biospecimen and Image sharing initiative we have been uploading and deidentifying images obtained over 25 years to our Image Analytics Platform, XNAT. This has been challenging due to the disparate formats of the raw images and the need to create custom scripts to remove identifying information from the image headers. We have uploaded most of our images and once complete, we will leverage this platform for image sharing.

We have recently started imaging sessions for the Bioimaging and Fluid Biomarkers core. We have constructed REDCap forms to capture participant information, imaging data and specimen tracking. These data will be incorporated into our centralized database and once we have a significant amount of participant data, these will also be made available through our Data Discovery Tool. We anticipate new efforts in the coming year to collect more biospecimens and images and we will assist in creating the necessary REDCap instruments and reporting from our centralized database.

#### **Proposed One-Year and Long-Term Outcomes:**

We anticipate that by focusing on building infrastructure to share Data, Images and Samples from our affiliated cores we will enable further research into Alzheimer’s Disease. Additionally, with the start of the Bioimaging and Biofluids core and other imaging and biospecimen collection efforts we will be able to incorporate these data into our centralized database.

### **Year End Progress Summary:**

We continue to build out our IT infrastructure to support our various programs which includes expanding our Data Discovery tool to include data from both our APOE Gene Dosing and ADCC programs as well as adding a feature where consortium members can download de-identified data. We have increased the number of reports available through our reporting portal to over 20; including one recently piloted that is used at the Diagnostic Consensus Conference to assist in reviewing participant history rather than having to go through paper charts. A new feature has been added to the report portal to enable users to subscribe to reports which are emailed on a monthly basis. Additionally, numerous new Tableau reports have been created and are distributed and reviewed at regular meetings for our ADCC, APOE and BIFB meetings. The Recruitment and Retention forms which were deployed last year have been adopted and populated by sites and we have generated a tableau report from these to help inform recruitment strategies. We have integrated NACC export tool, Issue Tracker and Reporting applications into a single code base, so users do not have to go to different locations while implementing granular permissions throughout. With respect to our data submissions to NACC, we have continued to reduce time to finalization by working with consortium partners to optimize workflows at each site.

We executed a three-way contract between Mayo Clinic, Banner Health and USC/GAAIN to be able to deposit data from our APOE gene dosing program on the GAAIN platform which permits investigators to run summary statistics over the data to determine its value for research projects. We have finalized our data dictionary with GAAIN and are ready to send data once GAAIN has completed their IRB renewal. Once the data is uploaded, GAAIN will have a link to our Data Discovery Tool which permits investigators to request data, with a robust tracking mechanism.

The upload and documenting of our historical images to our image analysis platform, XNAT, has been completed. All newly acquired images are automatically de-identified and uploaded to XNAT. Having cataloged all our images as well as our samples and building out our centralized database, permitted us to share data, images and samples on 567 participants with collaborators.

During the current reporting period, we went live with our REDCap BIFB project and participated in bi-weekly operations meeting to get feedback and make changes to better support the operations of the program. Several Tableau reports have been made to monitor the enrollment in this program, tracking consent, biospecimen and image acquisition.

Additional progress outside of the stated aims include: Optimization of submitting data to NACC from BSHRI's system by writing a program which only sends data on non-finalized packets; creation of an application to import all finalized NACC data into Banner's Teradata Database system for ad-hoc reporting requests; added functionality to our reporting portal to accommodate the upload and distribution of Tableau reports; added functionality to our NACC export tool, to enable the export of a single participant/visit.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Statistical and Neuroimaging Data Science Core Resources Serving the Consortium Members.** Yi Su, PhD, Dhruvan Goradia, PhD, Hillary Protas, PhD, Michael Malek-Ahmadi, PhD, Yinghua Chen, MS, Ji Luo, MS, Wendy Lee, MS, Gene Alexander, PhD, Rui Chang, PhD, Amylou Dueck, PhD, Ben Readhead, MBBS, Don Saner, MS, Kewei Chen, PhD, Eric M. Reiman, MD. Banner Alzheimer's Institute; Arizona State University; Banner Sun Health Research Institute; Mayo Clinic Arizona; Translational Genomics Research Institute; University of Arizona; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) To generate and facilitate the sharing of imaging-based measurements generated from state-of-the-art methodologies to leverage multiple large datasets, such as Arizona APOE study, ADNI, and others, to allow combined analysis for improved power and sensitivity.
- 2) To offer comprehensive statistical and image analysis services to investigators in the Consortium and local AD research community based on their needs.
- 3) To maintain and establish collaborative research within the local statistical and data science research community.

### **Background and Significance:**

AD is a disorder with complex etiology and multiple pathogenesis pathways, and currently, there are no disease-modifying therapies for AD, although a variety of treatment strategies are being explored. The development of effective interventions to prevent or delay the onset of AD requires an in-depth understanding of the underlying mechanisms that lead to neurodegenerative changes, which in turn result in cognitive decline and dementia. Such efforts are greatly enhanced by the availability of comprehensive dataset such as ADNI, with imaging, fluid biomarkers, cognitive, and clinical measurements. A major local effort within the Consortium is the long running Arizona APOE study which has collected neuroimaging data on over 400 participants with more than 1400 magnetic resonance imaging (MRI) scans and close to 2000 positron emission tomography (PET) scans that characterize the structural, metabolic, and pathological changes of the brain. This dataset is however underutilized hampered in part due to our limited capacity to perform customized analysis for interested investigators. A standardized set of imaging variables derived following well established state-of-the-art methodologies that are widely used would greatly improve the efficiency of the data analysis, availability of the dataset, and reduce the redundancy in repeated reanalysis with small changes in analysis procedures. It also can leverage the availability of other public datasets such as ADNI and allow combined analysis with larger sample size and improved statistical power. Over the years, the CIAL team has continued to serve as a core resource of imaging and statistical expertise to facilitate AD and neuroimaging research by collaborating with local, national, and international investigators. With state funding support through the Arizona Alzheimer's Consortium, our lab has helped collaborating investigators perform imaging and statistical analysis using state-of-the-art methodologies developed by our lab and elsewhere. The important research and analysis performed through this grant has the potential for publication and lasting impact on the general research field that would otherwise lack funding support, with a few examples from the previous year listed here. The lab has also helped collaborating investigators with preliminary data analysis, neuroimaging study design and statistical power analysis to facilitate their grant applications in the planning phases and participate as part of their research team if funded. We would like to continue this effort and keep supporting the research and development of Consortium investigators. The Consortium

undergone some personnel changes in the past year especially in the statistical and data analysis team. Dr. Yi Su was recruited to take leadership role of the CIAL team and the addition of Dr. Ben Readhead (ASU) and Dr. Rui Chang (UA) who are experts in bioinformatics and big data analysis. In conjunction with support from the NIH funded ADCC DMSC, we would like to further develop and maintain a strong statistical and data analysis team for the Consortium by continue to work with established team members such as Dr. Alexander (UA) and Dr. Dueck (Mayo) and establish regular meetings to discuss ongoing collaborations and new data analysis needs. We are glad to report that Dr. Kewei Chen is back at BAI full time where he can devote more of his time and efforts into research and collaborations.

### **Experimental Designs and Methods:**

Aim 1. We will apply the standardized imaging analysis procedures on available dataset to generate a database of imaging derived measurements of regional amyloid and tau burden, global amyloid burden index, regional glucose metabolic rate, the HCl, regional volumetric measurements, and make them available to collaborators. We will also investigate and identify the common variables that can be derived from the Arizona APOE cohort, ADNI, and Open Access Series of Imaging Studies (OASIS) Brain (<https://www.oasis-brains.org>) datasets (OASIS-3 in particular).

Aim 2. We will continue our effort to facilitate AD research by providing data access in addition to imaging and statistical expertise.

Aim 3. We will work with Dr. Alexander on the development of standardized image and statistical analysis procedures to facilitate the effort on bioimaging-fluid biomarker project. We will also work with Drs. Readhead and Chang on the development of strategies of combined imaging and omics data analysis and provide expertise on big data analysis to the local research community. We will also establish regular meetings to discuss ongoing collaborations.

### **Proposed One-Year and Long-Term Outcomes:**

In the upcoming year, we will generate and curate a standardized imaging dataset based on the Arizona APOE cohort and make it available to collaborators. We anticipate high quality publications and grant submissions as part of the collaborative effort. In long term, we anticipate being able to help grow the research portfolio of Consortium investigators through our service and assistance.

### **Year End Progress Summary:**

In the past year, the Computational Image Analysis Laboratory (CIAL) continued to serve as a core resource for the consortium and local, national, and international investigators to facilitate sharing of data and expertise in imaging and statistical methodology. We generated and curated imaging derived measurements using standardized pipelines developed in the lab and commonly used imaging analysis tools by the broad AD research community. Based on the Arizona APOE cohort, we derived FreeSurfer based volumetric and cortical thickness measurements on 1422 T1-weighted MR scans, PUP based regional relative glucose metabolic rate measurements from 1312 FDG PET scans, PUP based regional amyloid burden measurements from 264 PiB PET scans and 117 florbetapir scans, and PUP based regional tau burden measurements from 142 flortaucipir scans. Using SPM based analyses, 1569 hypometabolic convergence index (HCI) measurements, 266 PiB SUVRs, 122 florbetapir SUVRs, and IPCA based whole brain shrinkage measure on 70 participants were also available. From the ADNI cohort, we have also curated SPM based results with 3646 HCIs, 3339 sROI measures, 2697 florbetapir SUVRs, 618 Tau SUVRs, and IPCA based whole brain shrinkage measure on 340 participants with associated demographic and clinical assessment data. From the OASIS-3 database, we curated FreeSurfer

based measurements from 2047 T1-MRIs, PUP based amyloid measurements from 935 PiB PET scans and 417 florbetapir scans with associated demographic and clinical assessments. These data are available to share with consortium members and collaborators. We are also currently generating standardized measurements of FLAIR based white matter hyperintensity measurements as well as additional imaging derived measurements from other available imaging modalities including rs-fMRI and DTI.

With state funding support through the Arizona Alzheimer's Consortium, our lab continued to help collaborating investigators perform imaging and statistical analysis using state-of-the-art methodologies developed by our lab and elsewhere. The important research and analysis performed through this grant has the potential for publication and lasting impact on the general research field that would otherwise lack funding support. The lab has also helped collaborating investigators with preliminary data analysis, neuroimaging study design and statistical power analysis to facilitate their grant applications in the planning phases and participate as part of their research team if funded. We continued to work with Dr. Caselli on the comparison between MCI converters ( $n = 65$ ) and nonconverters ( $n = 719$ ) in the Arizona APOE cohort, 34 of 35 cognitive measures and 9 of 18 behavioral measures declined faster post-inflection in the MCI converters determined based on piece-wise linear mixed effect models; the earliest cognitive inflection point was nearly 20 years in advance of MCI diagnosis, suggesting the preclinical duration of cognitive and behavioral changes approaches the earliest reported biomarker changes. Additional analysis with imaging biomarkers on a subset of this cohort is ongoing (Alzheimer's & Dementia 2019). In collaboration with Dr. Jun at the Boston University and a group of investigators in the Alzheimer's Disease Genetics Consortium, we reported that APOE2 homozygotes had a 99.6% lower risk in developing AD than APOE4 homozygotes based on a large neuropathologically characterized cohort ( $>5000$ ) (Nature Communication 2020). We also reported in a case study that an ADAD mutation carrier with a rare variant of the APOE gene, the Christchurch mutation, in homozygotes configuration did not develop cognitive symptoms until 70s, whose imaging data suggests a high amyloid load but low tau burden and relatively normal glucose metabolism (Nature Medicine 2019). Based on these findings and additional research from the NIH APOE study (R01AG031581), we submitted a new R01 application to expand the APOE study under the leadership of Dr. Reiman. Working with Dr. Yalin Wang (ASU), we continued to support his methodological development by providing high quality data curated from the Arizona APOE cohort, ADNI, and more recently OASIS-3 (Human Brain Mapping 2019, Neuroimage Clinical 2019). We also participated in two NIH grant applications Dr. Wang submitted, and one of them received a fundable score (14%) recently. We also continue to work with Drs. Braiden (ASU), Schaefer (ASU), Beach (BSHRI), Atri (BSHRI) and his visiting scholar from Czech Republic to support their research and preparation of scientific publications.

This state funded Statistics and Neuroimaging core resources also continued to work with the Data Management and Statistics Core (DMSC) of the NIH funded Alzheimer's Core Center (P30AG019610) to collaborate with core investigators and serve regional research communities. We are currently working with Dr. Alexander (UA) to examine white matter changes in the Arizona APOE cohort in relationship to aging and APOE genotype. We are also currently investigating the relationship between imaging derived biomarkers with metabolomics profiles using data from the ADNI cohort in collaboration with Dr. Chang (UA). Partially supported by this State funding, we take the lead on the Data Management Core and the Statistical Core for the NIH funded Alzheimer's Core Center program by organizing monthly meetings, reaching out to consortium-wide investigators for their statistical and machine learning needs, and organizing statistical seminar and conferences.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Enhancement of Native American Recruitment and Enrollment.** Eric Reiman, MD, David Coon, PhD, Richard Caselli, MD, Thomas Beach, MD, PhD, Lori Nisson, LCSW, Edward Zamrini, MD, David Weidman, MD, Meredith Wicklund, MD, Geoffrey Ahern, MD, Steven Rapcsak, MD. Banner Alzheimer's Institute; Mayo Clinic Arizona; Banner Sun Health Research Institute; Barrow Neurological Institute; University of Arizona; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) Support specific efforts to increase enrollment of Native Americans into the ADC through collaborations with other research studies and/or registries that have Native American participants who have agreed to be re-contacted about other research studies.
- 2) Consult with research study team members mentioned in Specific Aim 1 to identify successful recruitment and retention efforts for Native American participants in their studies.

### **Background and Significance:**

This proposal requests complementary support to enhance ongoing enrollment efforts of Native Americans as part of the Arizona Alzheimer's Research Consortium's ADC and ancillary programs. The Arizona ADC is part of a multi-institutional statewide consortium that links together the major research institutions in Arizona to advance effort in the early detection, tracking of progression, and evaluation treatments and prevention therapies for Alzheimer's disease and related disorders. The ancillary programs include the Arizona BBDP and Arizona APOE4 Gene Dose Program. The Arizona Brain and Body Donation Program (BBDP) provides an invaluable scientific resource of longitudinal cognitive, motor, clinical, and genetic data from >800 living older adults who have standardized annual assessments, consent to brain (and frequently body) donation, and provide a resource of unusually high-quality brain tissue, postmortem CSF and blood samples (which differ in some respects to samples that are acquired in life) and neuropathological data after they die. The program includes but is not limited to research participants with the clinical features of Alzheimer's disease (AD) or related disorders and cognitively and neurologically unimpaired older adults with support from the National Institute on Aging (NIA)-supported Arizona AD Core Center (ADCC), research participants with the clinical features of Parkinson's disease (PD) and related disorders and cognitively and neurologically unimpaired older adults with support from the National Institute of Neurological Disorders (NINDS)-supported National Brain and Tissue Resource for PD and Related Disorders (NBTR-PD). The Arizona APOE4 Gene Dose Program provides an invaluable scientific resource of longitudinal data from initially cognitively unimpaired research participants with two, one and no copies of the APOE4 allele, the major genetic risk factor for AD. The program includes nearly 200 participants who were initially late-middle-aged participants with a first degree family history of dementia who are followed every two years with a battery of clinical ratings, cognitive tests, FDG, amyloid and now tau PET scans, and MRIs, who have provided plasma, serum and PBMC samples that are stored at Mayo Clinic, and who have begun to provide CSF samples with support from a longstanding NIA grant. It also includes more than 200 other participants, with or without a family history and through youngest to oldest adult ages, who are followed using state and organizational Arizona Alzheimer's Consortium funds, and who have not yet provided CSF, plasma and serum samples.

**Native American Enrollment:** The inclusion of participants with Native American characteristics will assist investigators in providing answers to questions about dementia diagnosis, treatment, and management strategies that are likely to be applicable to the broader Native American population in the U.S. Additionally, a more diverse participant pool that includes Native Americans will facilitate investigations of different risk factors, health disparities and the neuropathology and genetics of AD and related dementias as well as studies of caregiving and family burden in diverse groups.

**Preliminary Data and Plan:**

We will collaborate with research study investigators who have been successful in enrolling and retaining Native American participants (e.g., Strong Heart NIH study) and implement a plan to enroll as many as possible of their Native American participants age 50 and older who agreed to be re-contacted about additional research opportunities. We will consult with study team members about other successful recruitment and retention strategies for Native American populations.

**Proposed One-Year and Long-Term Outcomes:**

The proposed outcomes would be to increase enrollment and retention of Native American participants into the ADCC and its ancillary programs. We expect to increase Native American enrollment by approximately 30 participants.

Funds will be used in a way that complement but do not overlap with funding provided by the NIA and other funding sources related to outreach, recruitment, and retention efforts with ADC participants.

**Year End Progress Summary:**

Our current Native American enrollment totals 34 active participants including 30 from the Clinical Core (26 active, 4 pending), 4 from our NIH Longitudinal Cohort, and 1 from our Brain and Body Donation Program.

During this reporting period, we made great strides in Native American outreach including establishing a partnership with MedStar Health Research Institute who implement the Strong Heart Study in the Phoenix Metro Area. Through the partnership, Medstar engages with Native Americans and informs them about the ADCC. Interested participants are then scheduled for enrollment visits at BAI. This partnership has led to 30 referrals, 18 scheduled appointments, and 4 enrollments. Unfortunately, in-person research visits have been postponed at this the so the scheduled were cancelled. We are currently making plans to complete telephonic enrollments of these participants.

The relationship with Medstar allowed us to overcome one of the common barriers associated with enrolling the Native American population. Often, participants do not show for their appointments because they have difficulty securing transportation. Medstar staff provide transportation for the participants to their appointments. It is worth nothing that this is the same staff member involved in the initial engagement. This continuity is likely to aid in retention long-term.

**Project Progress Reports**  
**Banner Sun Health Research Institute**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Developing a Shared Resource of Cerebrospinal Fluid, Plasma, Serum, and Peripheral Blood Mononuclear Cell (PBMC) Samples from Arizona's Longitudinal Brain and Body Donation and Apolipoprotein E4 (APOE4) Gene Dose Programs.** Thomas G. Beach, MD, PhD, Edward Zamrini, MD, Geidy Serrano, PhD, Kathryn Demarco, David Weidman, MD, Lucia Sue, Richard J. Caselli, MD, Charles H. Adler, Donald Saner, Eric M. Reiman, MD. Banner Sun Health Research Institute; Banner Alzheimer's Institute; Mayo Clinic Arizona; University of Arizona; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1. To develop a repository of cerebrospinal fluid (CSF), plasma, and PBMC samples, linked to brain imaging and neuropathology data from well characterized, longitudinally assessed, and consenting participants in Arizona's Brain and Body Donation Program.

2. To develop a repository of CSF, plasma, and PBMC samples linked to brain imaging and neuropathology data from well characterized, longitudinally assessed, and consenting participants in Arizona's APOE4 Gene Dose Program.

3. To provide a shared resource of CSF, plasma, and PBMC samples and data linked to brain imaging and neuropathology data to researchers inside Arizona and around the world.

**Background and Significance:** The Arizona Brain and Body Donation Program (BBDP) provides an invaluable scientific resource of longitudinal cognitive, motor, clinical, and genetic data from >900 living older adults who have standardized annual assessments, consent to brain (and frequently body) donation, and provide a resource of unusually high-quality brain tissue, postmortem CSF and blood samples (which differ in some respects to samples that are acquired in life) and neuropathological data after they die. The program includes but is not limited to research participants with the clinical features of Alzheimer's disease (AD) or related disorders and cognitively and neurologically unimpaired older adults with support from the National Institute on Aging (NIA)-supported Arizona AD Core Center (ADCC), research participants with the clinical features of Parkinson's disease (PD) and related disorders and cognitively and neurologically unimpaired older adults. The BBDP has provided an invaluable resource of data, brain tissue and DNA to researchers around the world. CSF and blood samples would enhance the value of the BBDP in several ways, including a) the chance to clarify whether the participants have CSF evidence of amyloid- $\beta$  and tau pathology (biomarkers of AD), b) the chance to evaluate, further develop emerging CSF and blood-based biomarkers in terms of the extent to which they predict subsequent clinical decline and the neuropathological diagnosis of AD, PD, and other disorders, and c) the chance to use CSF- and blood-based measurements to further help in the clarification of disease mechanisms and risk factors. This year the project will include brain imaging of a subset of subjects, with MR, amyloid PET and tau PET, allowing cross-comparison of multiple AD biomarkers.

The Arizona APOE4 Gene Dose Program provides an invaluable scientific resource of longitudinal data from initially cognitively unimpaired research participants with two, one and no copies of the APOE4 allele, the major genetic risk factor for AD. The program includes nearly 200 participants who were initially late-middle-aged participants with a first degree family history of dementia who are followed every two years with a battery of clinical ratings, cognitive tests, FDG, amyloid and now tau PET scans, and MRIs, who have provided plasma, serum and PBMC samples that are stored at Mayo Clinic, and who have begun to provide CSF samples with support

from a longstanding NIA grant. It also includes more than 200 other participants, with or without a family history and through youngest to oldest adult ages, who are followed using state and organizational Arizona Alzheimer's Consortium funds, and who have not yet provided CSF, plasma and serum samples. CSF and blood samples in state-supported APOE4 Gene Dose participants would increase the value of the Arizona APOE4 Gene Dose Program in several ways, including a) the chance to detect and track the earliest fluid biomarker changes associated with the predisposition to AD, b) clarify the extent to which they are associated with subsequent cognitive decline and clinical progression, c) help to distinguish the cognitive changes associated with preclinical AD from those associated with aging in the absence of AD pathology, d) help researchers clarify the extent to which emerging AD biomarkers could be detected at earlier ages, and e) provide promising endophenotypes to help in the clarification of AD risk factors.

**Experimental Designs and Methods:** During the one-year funding period, we propose to further develop the infrastructure to conduct lumbar punctures (LPs), acquire up to 30 ml of CSF and 40 ml of blood and process CSF, plasma, and buffy coat (for PBMCs) samples from BBBDP participants at BSHRI and APOE4 Gene Dose Program participants at BAI, to process, aliquot and store samples using standardized procedures, and to establish a repository of these samples at BSHRI. This year, we propose, at a minimum, to acquire CSF samples in 30 returning BBBDP participants at BSHRI and in 20 returning state-supported APOE4 Gene Dose Program participants at BAI who consent to LPs; and we propose to acquire, at a minimum, blood samples in 400 returning BBBDP participants and in the 50 APOE4 Gene Dose Program participants. Additionally, we will perform brain imaging, to include MR, amyloid PET and tau PET, on 10 BBBDP subjects that also contribute blood and CSF. The biospecimens and imaging data from these 33 subjects will augment a further 24 subjects that will annually contribute, using identical protocols, blood, CSF and the same brain imaging scans, as part of a new supplemental grant from the National Institute on Aging to our Arizona Alzheimer's Disease Core Center (funding date started September 2018).

**CSF Samples.** LPs will be acquired by trained and experienced personnel standardized procedures established for other longitudinal cohorts. We propose to acquire up to 40 ml of CSF, which will then be centrifuged at 1,500 rpm for 10 min at 24°C. The supernatant will be collected, placed into 0.25 ml aliquots, and stored at -80°C. One (1) ml of CSF from each subject will be sent to a commercial lab for standard analyses on cell count, protein and glucose levels, and hemoglobin levels.

**Blood Samples.** We propose to acquire up to 50 ml of venous blood in EDTA tubes. Blood will be centrifuged at 1,500 rpm for 15 min at 24°C to separate plasma and red blood cells. The plasma will be collected and placed into 1.7 ml microcentrifuge tubes and then centrifuged again for 5 min, 4°C at 14,000 rpm. From blood samples collected at BSHRI, the buffy coat will be further refined using standard methodology to provide purified peripheral blood mononuclear cells (PBMC), which, along with the plasma aliquots, will be stored at -80°C.

**Fluid Repository.** All samples from Specific Aims 1 and 2 will be stored at BSHRI in ultra-low temperature freezers protected with redundant temperature-activated alerts, banks of emergency CO2 tanks, redundant air conditioning units and backup diesel alternate power supply. BBBDP staff are on constant call to respond to freezer alerts. A biological sample distribution committee involving the BBBDP and APOE4 Gene Dose Program PIs will evaluate all research proposals involving the use of shared biological samples.

## **Year End Progress Summary:**

### **Blood Samples**

To date since 2015, we have obtained stored in-house 27,519 blood samples from 742 participants on 1,488 different appointments, from which 130 already came to autopsy and 151 were collected this funding period (as of March 02, 2020). In addition, we collected 900 blood samples from 60 individuals who also received or who are eligible to receive MR, 18F Flortaucipir PET and 11C PiB PET scans, these samples are currently stored at NIH for further multi institutional collaborations. By clinical diagnosis, the collected blood samples are from 490 non-demented controls, 124 subjects with mild cognitive impairment, 60 subjects with a clinical diagnosis of dementia due to possible or probable Alzheimer's disease, 140 subjects with Parkinson's diseases and 49 with other diagnoses. It is projected by the end of the funding year 2019-2020 we will collect and save blood samples from 250 BBDP participants.

Thanks to this initiative this year we were able to share blood samples from individuals that already came to autopsy and have a final clinicopathological evaluation with six different scientist groups. Two of those scientist groups are from institutions in Arizona and four are out of state, continuing our tradition of open collaboration. These transferred samples have already resulted in a high impact publication in Nature Medicine and to our knowledge one additional important publication that is currently in preparation.

### **CSF Samples.**

To date since 2015, we have obtained 1,583 CSF samples from 44 cognitively normal participants, from which 5 already came to autopsy and 21 were collected in this funding period (as of March 02, 2020). In addition, we collected 280CSF samples from 21 cognitively normal individuals who were also received or who are eligible to receive the above-mentioned scans, these samples are currently stored at NIH for further multi institutional collaborations.

Fluid Repository. All samples from Specific Aims 1 and 2 are stored at BSHRI in ultra-low temperature freezers protected with redundant temperature-activated alerts, banks of emergency CO2 tanks, redundant air conditioning units and backup diesel alternate power supply. BBDP staff are on constant call to respond to freezer alerts. A biological sample distribution committee involving the BBPD and APOE4 Gene Dose Program PIs will evaluate all research proposals involving the use of shared biological samples.

## **References**

1: Janelidze S, Mattsson N, Palmqvist S, Smith R, **Beach TG**, **Serrano GE**, Chai X, Proctor NK, Eichenlaub U, Zetterberg H, Blennow K, **Reiman EM**, Stomrud E, Dage JL, Hansson O. Plasma P-tau181 in Alzheimer's disease: relationship to other biomarkers, differential diagnosis, neuropathology and longitudinal progression to Alzheimer's dementia. Nat Med. 2020 Mar;26(3):379-386.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**A Human Brain Single-Cell Suspension Resource.** Geidy Serrano, PhD, Thomas G. Beach, MD, PhD, Lih-Fen Lue, PhD, Matthew Huentelman, PhD, David Brafman, PhD. Banner Sun Health Research Institute; Banner Alzheimer's Institute; Mayo Clinic Arizona; University of Arizona; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1. Develop, optimize and standardize a method for producing single-cell suspensions from rapidly-autopsied human brains, allowing the analysis of proteins, RNA and DNA from single cells and phenotypically-specified cell populations.
2. Biochemically characterize single cells and phenotypically-specified populations of cells from single-cell suspensions created with an optimized standardized method (from Specific Aim 1).
3. To provide the foundation of a shared resource of separated cells to researchers within and outside Arizona.

**Background and Significance:** Biochemical analysis of human neurodegenerative brain tissue, especially from Alzheimer's disease (AD) and Parkinson's disease (PD) patients, has produced much of what is known about these conditions, and has led to the major FDA-approved therapies. The typical approach has been to homogenize whole pieces of brain tissue and separately characterize the proteins, RNA, DNA and other macromolecules within. While this has been sufficient to identify large changes, there is very little ability to identify small changes, or changes in small subsets of cells. Furthermore, neurodegenerative disease often leads to massive losses of the targeted and disease-relevant cells, for example the entorhinal cortex layer II stellate neurons or substantia nigra pigmented neurons. Whole-homogenate analysis of such brain regions can give completely misleading results, as any biochemical constituent that is selectively localized to the depleted cells will appear to be "down-regulated", whereas in fact it has most likely been lost only as an "innocent bystander". Also, a relevant loss or increase might be completely missed, if the biochemical entity is found in many cell types, diluting the "lost" signal from the cell of interest, especially if that cell type is uncommon or rare. To effectively investigate the biochemistry of neurodegenerative disease in the brain, with its thousands of different cell types, we must first separate the cells, and study them as phenotypically-defined populations and even as individuals. In recent years, methods have been developed that allow an initial creation of single-cell suspensions from solid tissue followed by analysis of phenotypically-defined cells sorted on the basis of cell-type identifying proteins or RNA expression. Some groups have already published intriguing results from AD brain cells, but as yet there has not been a comprehensive exploitation of these novel technologies utilizing postmortem human brain. This set of experienced neuroscience investigators, together with a unique rapid-autopsy brain tissue resource, are well-suited to apply these methods on a large scale to AD and other neurodegenerative brain diseases.

### **Experimental Designs and Methods:**

#### ***Creation of dissociated-cell suspensions***

In this project we developed a new method for the generation of whole-soma-dissociated-suspensions (WSDS) in fresh human brain that could be shared with scientists as a resource to study human single cells or cell types populations. WSDS were generated using a hypothermic

cell dissociation which aimed to minimize molecular changes caused by processing, by cold processing. The hypothermic approach uses enzymatic digestion with Accutase for 4 hours at 4°C in fresh tissue minced with a razor blade, followed by mechanically disrupted by repetitive pipetting. Myelin, neuropil and other cellular debris removal is done by using different densities of percoll and centrifugation.

To date, 262 autopsies have been performed by the Brain and Body Donation Program (BBDP) since the funding start date for this continuing project (July 1, 2016). Of these, tissue from 120 subjects has been used to generate WSDS. In this current funding year (beginning July 1, 2019), we have obtained cell suspensions using the hypothermic protocol from 17 autopsies out of 47 total autopsies. On average we are now collecting 10.0 million cells/gram of tissue. Final suspensions are aliquotted for tissue banking in cryopreservative solution and stored at -80°C and for quality control (QC) assessments.

### **Specific Aim 1: Methods paper preparation and additional funding.**

A methods manuscript was already written in collaboration with Matthew Huentelman's laboratory, the manuscript is under internal review and will be sent to Cell and Tissue Banking Journal before the end of this funding year. This manuscript describes the methodology used to generate WSDS from samples collected from rapid autopsy and thoroughly characterize the samples. This manuscript will be of high importance in the field, as many keep using single nuclei and single cells analyzes without reporting careful characterization of the cell or nuclei population and proper comparisons of the suspensions with the original source.

This funding year we applied for and received funding from the Michael J Fox Foundation (MJFF) with the goal of understanding the role of astrocytes in Parkinson's disease. Until now the vast majority of evidence of astrocytic dysfunction in aging and Lewy body disease (LBD) has come from experiments done with whole tissue homogenates, astrocytes collected by laser capture or cell cultures derived from animal models or cell lines. In the proposed study we will conduct an unbiased comparison of human astrocyte gene expression in PD, LBD and control subjects. We will do whole transcriptome sequencing from enriched human astrocyte populations sorted by fluorescence activated cell sorting (FACS) using anti-GFAP antibodies. We propose that the number of sequenced transcripts will thus likely be significantly greater using this approach than those obtained by groups using laser-capture. Single-cell suspensions will be prepared from fresh human cerebral cortex prospectively obtained at rapid autopsies from the Arizona Study of Aging and Neurodegenerative Disease (AZSAND)/Brain and Body Donation Program (BBDP).

### **Specific Aim 2: High-profile projects, to further establish the importance of the general approach and to further awareness of the resource among the neurodegenerative disease scientific community**

During this funding year we have done multiple FACS experiments with different antibodies aiming to improve cell enrichment. We have used multiple cell specific markers to allow us to sort different cell populations, but also used FACS to sort by proteins that are expressed in disease, such as phosphorylated tau (p-tau) and phosphorylated synuclein (p-syn). A total of six cases with Alzheimer's disease (AD) were sorted with p-tau antibody and five non demented controls were sorted with neurofilament in order to study possible neuron-specific changes in AD in response to the presence of the disease-causing altered proteins. RNA from the enriched populations was isolated and sent for RNA sequencing (RNA-Seq). Analysis results are pending. In addition, in order to generate pilot data for a grant with the Parkinson's disease association, we sorted cells containing phosphorylated alpha-synuclein ( p-syn) s from one case with Parkinson's disease (PD) and compared these with cells sorted from one movement control with a neuronal marker (NeuN). More than 11,000 gene transcripts were sequenced and compared between p-

syn and NeuN-sorted cell pools. Network enrichment analysis was remarkable for a 50-fold increased abundance of DAPK1 (death-associated protein kinase 1) and of calcium signaling pathways in general. DAPK1, like LRRK2, is a member of the ROCO kinase family and is thought to be a regulator of apoptosis and autophagy. It may act to stimulate the autophagic pathway through lysosomal and endoplasmic reticulum calcium release. DAPK1 modulators have been proposed for neurodegenerative disease therapies. Another interesting hit was for HSPA12A, a member of the HSP70 gene family, which is known to regulate protein misfolding in a variety of diseases, including  $\alpha$ -syn in PD and tau in AD, perhaps by channeling misfolded proteins for degradation by the proteasome. Amongst transcripts significantly less common in the p-syn-labeled cells was SNAP25, a membrane protein involved in synaptic vesicle fusion. These preliminary data are just a very early look but are encouraging for our approach at least in terms of the successful sequencing of large numbers of transcripts.

Another methodological approach that we are continuing to work on is to use the 10X Genomics droplet-based platform, using WSDS for single cell RNA-Seq. Challenges so far have been adapting the protocols used by our collaborators in the TGen and Mayo clinic laboratories. Dr. John Fryer from Mayo Clinic, whose laboratory is already proficient using 10x in whole cells from mouse brain, obtained WSDS from us to do scRNAseq from whole cells (rather than nuclei) which could potentially be the method that will give the greatest advances in the understanding of cell-type-specific gene expression changes. Unfortunately, this methodology seems not be appropriate for human WSDS because the cells appeared to be damaged by the process. We hypothesized that cell size and the neurite presence in the suspension is our major challenge with this approach. We are now trying new approaches that will be gentler approaches for single cell sequencing, such as manual bar coding using SplitBio technology. We have had multiple discussions with the scientist who developed this technique and currently have everything necessary to experiment with this approach. We are optimistic about the capabilities of this technique and think this will help us generate valuable data.

### **Year End Progress Summary:**

1. Over the last couple of years, we had collected enough evidence that show WSDS generated at the Banner BBDP are suitable for multiple experiments that could lead to better understanding of single cell or population changes in aging and neurodegenerative disorders associated with aging. We are now actively promoting this resource in our website and meetings.
2. During this year three more high-profile projects were undertaken to further establish the importance of the general approach and to further awareness of the resource among the neurodegenerative disease scientific community. We will soon have enough primary data that will allow us to publish additional manuscripts and request additional funding from government and private institutions.
3. Further analysis of cell suspensions using next generation RNA sequencing (RNA-Seq) will be done using gentler approaches such as manual bar coding using SplitBio technology and magnetic beads covered with a surface epoxy group for manual cell separation. Both approaches seem very promising and we are already in the process of testing the protocols.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Establishing human subject-derived fibroblasts and microglia cells core for translational research.** Lih-Fen Lue, PhD, David Brafman, PhD, Thomas G. Beach, MD, PhD, Geidy Serrano, PhD. Banner Sun Health Research Institute; Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) To bank and characterize human microglia for culture with standardized procedures.
- 2) To bank and characterize postmortem scalp-derived fibroblasts with standardized procedures.
- 3) To generate and bank ApoE genotype-specific iPSC lines from patients' scalp-derived fibroblasts

### **Background and Significance:**

Genetically engineered AD mouse models have played an important role in pointing to many key mechanisms in AD. However, transgenic mouse model-based research has not translated to effective human therapeutics [1-3]. The transgenic mice models lack the genetic, epigenetic, and transcriptomic heterogeneity present in human diseases [4;5] and are typically not representative of "sporadic" AD. Research in human subject-derived models is needed [7-13]. Currently, there are two types of human cell sources which have been used for producing human brain cell models: differentiated cells reprogrammed from induced-pluripotent stem cells (iPSC) and primary brain cells isolated directly from postmortem brains. To facilitate progress in human cell-based basic and translational research, it is crucial to ensure a continuous supply of primary cell cultures covering a broad range of neuropathologically-characterized brain heterogeneity [14]. This goal can only be achieved with a dedicated prospective program at a suitable center with rapid autopsy of large subject numbers as well as standardized clinical and postmortem research examinations. The Brain and Body Donation Program (BBDP) of the Banner Sun Health Research Institute (BSHRI), established since 1988, is situated at an advantageous position to provide both kinds of human cell resources due to a track-record of short postmortem delay (median = 3.0 hours) and a collection of medical, neurological and neuropathological data for each participant. The BBDP program, through the efforts of a well-established autopsy team, banks annually tissues from 60-90 cases of normal elderly subjects as well as patients with neurodegenerative diseases. Participants receive an annual standardized battery of clinical and neuropsychological assessment as well as comprehensive neuropathological assessment after death [15]. The program provides invaluable biomaterials for both academic and pharmaceutical communities. This proposal will take advantage of the BBDP at BSHRI to build a sustainable program for banking viable cells using postmortem brain and skin tissues. Our initial effort will be focusing on microglia and skin fibroblasts. Differentiation of microglia from iPSCs derived from other cell types has been a challenge [16], and there is a great demand for microglia directly isolated from patient's brain, to investigate inflammatory mechanisms and for anti-inflammatory drug development. Skin fibroblasts are the most accessible cell types that have been successfully made to iPSC or directly trans-differentiated to neural cells such as neurons and astrocytes. While fibroblasts are easy to obtain at many places, the advantage of using fibroblasts banked at BBDP is that they will be linked to deep phenotyping through associated neurological, neuropsychological, and histopathological data. The main objectives of the project are: (1) to standardize current protocols to obtain cells with consistent qualities; (2) to determine the viability and phenotypic features during storage of cryoprotected cells; (3) to determine basic gene and protein expression profiles of the cells produced from our standardized procedure; (4) to generate

ApoE genotype-specific hiPSCs from the banked fibroblasts. The success of the project relies on expertise and techniques in isolating neural and skin cells. The PI, Dr. Lue, has more than twenty years of experience in isolating human brain microglia [17-28]. She had also conducted skin fibroblast isolation from scalp tissues taken from 15 autopsy cases in a project previously funded by Arizona Alzheimer's Consortium. In last funding period she has banked scalp fibroblasts from 9 ApoE  $\epsilon$ 3/3 carriers and 3 ApoE  $\epsilon$ 3/4 carriers. Dr. Brafman is experienced in the development of iPSC cells, in last funding period he has started to make iPSC from the fibroblasts derived from  $\epsilon$ 3/3 and  $\epsilon$ 3/4 cases.

### **Experimental Designs and Methods:**

Specific Aim 1: This aim will (1) establish a shorter procedure for isolation of microglia, (2) characterize isolated cells with cell-type specific markers, (3) compare the effects of various cryogenic reagents on viability preservation during storage, and (4) bank cells. The isolation procedure for microglia will be based on our previously published procedure with further modification. The procedure requires 5 hours for isolation, followed by a 24-hour selective adherence procedure to obtain high purity of microglia. We will replace the selective adherence step by a magnetic beads-separation procedure using magnetic columns (less shear force). We will select a suitable cryoprotectant that gives a combination of highest survival rate and best RNA quality after cells are thawed from the frozen state.

Specific Aim 2: This aim will (1) modify the core procedure to increase the efficiency of fibroblast harvest; (2) characterize the cells using flow cytometry analysis; (3) assess revival rate in relation to type of cryoprotective reagents, and (4) bank cells. We have previously used a conventional procedure to isolate fibroblasts from epidermis of scalp tissues collected at autopsy. The procedure involves long-term in vitro expansion from primary epidermal and dermal cell mixed culture. We had used this procedure to isolate the fibroblasts from 15 cases in the past and 12 cases since September of 2018. One new goal has been added to this year's project; this is to test the cryoprotection of the tissues for cell isolation. If we could demonstrate that postmortem skin tissues could be cryoprotected and grow fibroblast after thawing, it will even expand our service to our research community by providing tissues for scientists to isolate their own fibroblasts.

Specific Aim 3: For generation of hiPSCs from patient fibroblasts we have adapted our previous published protocol for reprogramming patient skin fibroblasts (Cell Rep. 2014 Dec 11;9(5):1770-1780). Briefly, expanded fibroblasts will be transduced with 4 non-integrating sendai viruses each expressing Oct3/4, Sox2, Klf4, c-Myc. After 3 days, transduced cells will be plated onto mitotically arrested mouse embryonic fibroblast (MEF) feeder cells. HiPSC colonies will begin picked 15-21 days after transduction. Isolated hiPSC colonies will be cultured in hiPSC expansion medium (DMEM/F12 with 10% knockout serum replacement supplement with FGF2 and ascorbic acid). Loss of sendai virus will be determined after passage 10 by immunostaining with anti-sendai virus antibody. HiPSC lines will be analyzed for (i) characteristic hiPSC cell morphology, (ii) expression of pluripotency markers OCT4, NANOG, and SOX2 (iii) ability to differentiate in vitro populations representative of the three main germ layers, and (iv) a normal complement of 46 chromosomes.

### **Proposed One-Year and Long-Term Outcomes:**

We anticipate using standardized procedure to bank microglia and fibroblast, 20 autopsy cases from each cell type, during this funding period. We will also demonstrate the utility of our banked cells in 3-D cellular models.

## **Year End Progress Summary:**

### **A. Autopsy cases processed for fibroblast and microglia isolation:**

From July of 2019 to February of 2020, human cells core had processed postmortem scalp tissues from 22 autopsy cases, averaging one case every 1.5 week. This led up to a total of 44 cases since the start of this program in September of 2018. Nevertheless, not all postmortem scalp tissues were able to grow fibroblasts. Our record showed an overall success rate of 74%. Whether cells were grown out from scalp tissues could be determined by a combination of antemortem factors, as we have a standardized procedure for processing and culturing. We did find that AD cases had the lowest success rate at about 50%. Cases from normal controls and with PD diagnosis had comparable success rate at 87.5% and 89%, respectively. We determined whether Apolipoprotein (Apo) E genotype from donors affected the outcome of scalp tissue culture. Among the autopsy cases from which fibroblasts have been successfully grown, there were 2 subjects with ApoE  $\epsilon$ 3/4 genotype, one ApoE  $\epsilon$ 4/4 case, and 2 ApoE  $\epsilon$ 2/3 cases, while the rest of the cases were  $\epsilon$ 3/3. Our data did not support genotype effects from the cases that we have done. It will be assessed in the future whether a combination of  $\epsilon$ 4/4 and AD could be the reason for a lower success rate. We had also determined whether age, postmortem delay or cell processing delay, or pathological severity affected the outcome of the culture. Our analysis concluded that none of these were determinants for success of the fibroblast production. As for microglia, the cell yields ranged 1- 7 million cells per gram of tissues; the yields were significantly lower in the cases with neurological disorders combined than from NC cases ( $P < 0.01$ ). The microglia yields were not affected by genotypes, expired age of donors, or severity of AD or alpha synucleinopathy Lewy pathological features.

During this funding period, we have implemented two procedures in order to enhance the capability of the cell bank. First, starting January of 2020, we began to collect scalp tissues in cryoprotectant, so that we can extend the shelf life of the tissues. By doing this, we have options of prioritizing tissue processing for cases with gene mutations or ApoE  $\epsilon$ 4/4 carriers. By cryopreserving scalp tissues from every case, we also banked scalp tissues for future use. Second, in response to several inquiries of microglia subtypes, we have added microglia selection procedure using antibody-specific magnetic beads. We have tested CD11b as a pan microglia marker to purify microglia from the mixed glia population. We also tested the antibodies for microglia subtypes, such as TMEM 119 and TREM2. The selected microglia were cryoprotected and stored in liquid nitrogen. As microglia are present as heterogeneous populations in the brains, this approach will allow us to bank pre-selected microglia subtypes. We had processed 8 postmortem brain tissues for isolation of microglia from 5 AD, 1 MCI, 1 PD, and 1 PDD cases.

### **B. Characterization findings:**

We have optimized a panel of the fibroblast markers for the characterization of the banked cells. We used the techniques of immunofluorescence and western blotting. The fibroblasts banked are assessed by immunoreactivity on western blots for their positive expressions of fibroblast surface protein, fibroblast activation protein, Vimentin, CD73, CD105, PDGFRB, and Fibronectin. Our banked fibroblasts were negative for epithelial cell marker. Ongoing work for characterization is to detect the same marker panel at mRNA expression level.

### **C. hiPSC induction:**

Cells from selected cases with 3/3 and 3/4 ApoE genotypes were sent to Dr. Brafman at ASU for hiPSC induction as proposed in Aim 3. The progress according to Dr. Brafman: (1) Case 1854 was successfully reprogrammed. They have been fully characterized for their karyotype, expression of pluripotency markers, and tri-lineage differentiation. (2) Case 1857 did not reprogram as the fibroblasts did not grow out very well and could have interfered with reprogramming. (3) Case 1858 fibroblasts grew up and reprogrammed but individual clones did not grow out. (4) Case 1863 had not been done yet.

**D. Progress Summary:**

We have successfully established the procedures for routine isolation of scalp fibroblasts and microglia from autopsy cases. We have established a panel of the characteristic markers for each cell type. From two cases performed for iPSC induction by ASU collaborator, several clones had been obtained. Fibroblasts which have been stored in cryoprotectant are now available for AAC research scientists and scientists throughout the national and international communities. We proposed to bank cryoprotected fibroblasts from 15-20 autopsy cases this year; we will exceed this number by the end of June. During this funding period, we have also supported scientists who applied for research grants from NIH with letter of support and private foundation as Co-investigator. A manuscript for this work is in preparation.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Laying the Groundwork for Prodromal Lewy Body Dementia Research in REM Sleep Behavior Disorder.** David Shprecher, DO, MSci, Thomas G. Beach, MD, PhD, Charles H. Adler, MD, PhD, Shyamal H. Mehta, MD, PhD, Holly Shill, MD, Nan Zhang, MS, Joyce Lee-Iannotti, MD. Banner Sun Health Research Institute; Mayo Clinic Arizona; Barrow Neurological Institute; Banner University Medical Center-Phoenix; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) To enroll subjects with polysomnogram-confirmed RBD into the Banner Sun Health Research Institute Brain and Body Donation Program, a longitudinal clinicopathological study of normal aging and neurodegenerative disease in Sun City, Arizona.
- 2) To determine feasibility of using the Sleep Profiler device in place of gold standard sleep laboratory polysomnogram to diagnose RBD in a patient's home.

### **Background and Significance:**

Idiopathic REM sleep behavior disorder (iRBD) is a harbinger of neurodegenerative disease in the elderly.<sup>1</sup> A definite diagnosis requires the presence of dream enactment behavior, absence of a secondary cause (such as medications, brainstem lesions in tracts mediating REM atonia, or neurodegenerative disease) and polysomnogram (PSG) confirmation (demonstrating REM atonia and the absence of an RBD mimic such as nocturnal frontal lobe epilepsy or arousals related to sleep apnea.)<sup>2</sup> Over the last 15 years, evidence from multiple research groups world-wide has indicated that over 50% of those with iRBD will develop either parkinsonism or dementia within 10 years, with 80% or more converting after 20 years.<sup>3-5</sup> Autopsy studies have shown that the great majority of those dying with RBD have a brain disorder characterized by the accumulation of a protein called alpha-synuclein and are hence termed "synucleinopathies."<sup>6</sup> The major synucleinopathies are Parkinson's disease (PD) and dementia with Lewy bodies (DLB). To address the need for tissue-based biomarkers to explain and measure the risk of phenoconversion from iRBD to PD or DLB, we began (with recent AAC support) initial recruitment of individuals with iRBD into the BBDP during the previous funding period. Furthermore, we began a pilot study to validate a home device to improve efficiency and cost of iRBD diagnosis.

### **Experimental Designs and Methods:**

Specific Aim 1: To enroll subjects with polysomnogram (PSG)-confirmed RBD into the Banner Sun Health Research Institute Brain and Body Donation Program, a longitudinal clinicopathological study of normal aging and neurodegenerative disease in Sun City, Arizona.

Specific Aim 2: To determine feasibility of using the Sleep Profiler device in place of gold standard sleep laboratory polysomnogram to diagnose RBD in a patient's home. All subjects undergoing research PSG will also be consented to wear the Sleep Profiler device, paired with the Night Shift device (a wireless actigraphy device) to provide wireless data about limb movements. Sleep Profiler data will be independently reviewed by a sleep medicine physician blinded to subjects' sleep laboratory PSG findings.

### **Year End Progress Summary:**

*There were unanticipated delays in recruitment due to changes in staffing which are expected to improve efficiency in the long run. Another factor was low referral rate, which we have addressed by increasing our newspaper advertising expenditures. We have also given several outreach talks on PD and DLB research throughout the Phoenix valley, highlighting this study. Furthermore, we*

are working closely with our sleep medicine clinic at Banner University Medical Center to engender direct referrals of RBD patients for this study. We recently published our survey study results on RBD prevalence in Sun City, AZ (as well as a previously completed study on prevalence of RBD and other PD risk factors in Salt Lake City, UT).

### **Proposed One-Year and Long-Term Outcomes:**

At the end of year one, we expect to:

- 1) Complete research PSG/Sleep Profiler testing on at least 20 pRBD subjects and increase the total number of enrolled BBDP participants with iRBD to at least 15 individuals.  
*We have screened a total of 28 pRBD subjects to date, 7 within the 2019-20 funding period. A total of 16 subjects to date (6 within the 2019-20 funding period) have had research PSG. We have one research PSG scheduled and expect to complete a total of 15 research PSGs (over the 2019-2020 funding period). Total BBDP participants with PSG confirmed iRBD is now at 6.*
- 2) Begin participation as a site under subcontract for NIH award R34AG056639, "Neuroprotective treatment trial planning in REM sleep behavior disorder," PI Jo-Ei Yu (Washington University, St. Louis.)  
*We have officially completed our subaward contract and IRB approval; first subjects to be enrolled by end of March 2020 (with a target of 20 by 4/2021). In order to work out logistics of obtaining and entering data for the different protocols, we will now prioritize enrollment into the NIH subaward study first (then invite participants to co-enroll in the BBDP in later 2020.)*
- 3) Publish our pilot study comparing the home Sleep Profiler device to in-laboratory polysomnogram.  
*We presented our pilot data at four separate academic meetings, and are planning joint manuscript preparation (in collaboration with colleagues at Mayo Rochester).*

During the subsequent year, we expect to:

- 1) Arrange sustainable funding for the continued enrollment of iRBD subjects into our BBDP cohort. Establishment of a cohort of prospectively-assessed RBD subjects will be used as preliminary data to obtain NIH, PCORI, and/or Michael J. Fox Foundation grants to enlarge the cohort, conduct directed studies of clinical progression biomarkers, and potentially begin prevention trials to slow or stop progression to PD or DLB. As the principal investigators have a long-established record of obtaining federal and non-federal out-of-state funding, this project has a high probability of leading to larger, long-term state revenue inflow and increasing local employment.
- 2) Partner with the pharmaceutical industry to validate a wearable device to anticipate phenoconversion from iRBD to manifest neurodegenerative disease. We are currently a Parkinson Study Group site for a study sponsored by Biogen validating biometric wearables in newly diagnosed PD, and have had initial conversations about expanding this research to the iRBD population. Biogen is one of several companies developing alpha-synuclein antibody therapies for PD and related disorders.  
*We will be a partner site under a collaborative grant application (soon to be submitted by Yo-Ei Ju at Washington, St. Louis) to expand the current NIH funded prodromal synucleinopathy cohort study to include longitudinal follow up with novel biomarkers including neuroimaging, tissue, and biometric/wearable data. This will be submitted in response to an NIH U19 RFA: <https://grants.nih.gov/grants/guide/rfa-files/RFA-AG-21-013.html>*

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Enhancing Clinical and Biological Characterization of The Longevity Cohort Study: Global Staging and Biospecimen Banking.** Alireza Atri, MD, PhD, Kathy O'Connor, MS, Christi Belden, PsyD, Edward Zamrini, MD, David W Coon, PhD, Jessica J Powell, PsyD, Briana Auman, PsyD, Geidy Serrano PhD, Thomas Beach, MD, PhD, Michael Malek-Ahmadi PhD. Banner Sun Health Research Institute; Arizona State University; Northwestern University; Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1) Clinical research phenotyping, via global staging of participants, through use of an algorithm to identify and administer the Clinical Dementia Rating (CDR) scale assessment to study participants and their study partner (not previously required in LCS) in individuals at higher risk of cognitive impairment and dementia (CID);

2) Blood collection and biospecimen banking of 30 cc of plasma from study participants to be made available for collaborative research opportunities and for preliminary data investigations for grant applications.

### **Background and Significance:**

Now in its 13th year, "The Longevity Study: Learning from Our Elders Cohort" (LCS) is a unique Phoenix metropolitan area-based longitudinal study of psychosocial dynamics, lifestyle, physical activity, and cognitive function in successful aging of independent, community-dwelling, older individuals. The average age of participants is >80; and 40% of active subjects are ≥ 85, the oldest old, a rapidly growing demographic in AZ and the U.S. in whom there is a paucity of data regarding bio, and psychosocial factors associated with successful aging. Since its inception, the LCS has enrolled (as of 4/17/19) 1398 subjects, and currently follows 638 participants who undergo annual interviews and assessments, performed either at the BSHRI Center for Healthy Aging or in the participants' residences. The study captures a wealth of sociodemographic, medical, cognitive, physical, personality, lifestyle, and psychosocial data; and contains a strong portfolio of psychosocial variables with wide breadth and depth that are yielding publications (e.g. Refs 1-10 – for Refs see original grant application). However, its potential for future impact and funding is limited by a lack of clinical research characterization of the global status of participants (e.g. via a process such as the Clinical Dementia Rating, CDR, assessment), and by the availability of biospecimens to conduct collaborative and cutting-edge research to link bio- and psychosocial factors in the study of successful aging in the older, and the oldest old. As active participants age there is likelihood of developing impairments and a need to rigorously identify and characterize participants to better discern patterns of successful aging, and detect subclinical cognitive impairment and (very mild) dementia (CID). Additionally, with the advent of blood-based biomarkers and polygenic risk algorithms for Alzheimer's disease (AD) and CID available in the next few years, there is a need for biologic data that can be readily linked to psychosocial and cognitive variables collected in the LCS. This proposal aims to obtain important clinical phenotyping (staging) and bio- characterization data that will substantially enhance potential for biopsychosocial collaborative research, particularly related to cognitive resilience and reserve (see Refs 11-14), funding prospects and impact of the LCS.

Availability of these additional data and biospecimens will create a synergistic effect of adding value and impact potential for an enhanced LCS database to possess a complete range of quality biopsychosocial data in a unique population, the older and the oldest old. The expanded dataset

will provide a valuable resource that will be further leveraged to better understand biopsychosocial factors, their inter-relations, and their dynamics that are associated with successful aging, neural resistance and cognitive and functional resilience and reserve. Finally, validating clinical phenotypes, by assessing the global status of LCS participants, will improve recruitment of cognitively unimpaired subjects into the Brain and Body Donation Program (BBDP) and dual-enrollment. Increasing efficiency and quantity of dual enrollment between LCS and BBDP would serve to support continued enrollment of cognitively normal elderly, particularly the oldest old, into the BBDP, would provide critical cross-validation between these programs, and would allow additional opportunities for exciting and impactful science to be undertaken in the subset of dually-enrolled participants who will be highly characterized by psychometric, bio, clinical, psychosocial, and, ultimately, pathological data.

### **Experimental Designs and Methods:**

Aim 1: Clinical Phenotyping - Newly enrolled participants will be required to have a study informant; both will undergo CDR by a certified rater (~1-1.5 hours). In consideration for attrition, will enroll ~100 new participants (~30/year expected rate; additional attrition from new requirement for informant that some participants will not meet). Additionally, active participants deemed to be at higher risk of cognitive impairment will be identified via an algorithm, that includes risk factors, self or informant report, MoCA score of <26 at baseline, or a  $\geq 2$  point drop in MoCA from any previous score, to undergo CDR assessment (N~180-200). Finally, will obtain CDR of ~120 active participants dual-enrolled in LCS and BBDP. All participants will be assigned a global stage (e.g. cognitively unimpaired, subjective cognitive decline, MCI, mild dementia) based algorithm criteria or the CDR (N~400).

Aim 2: Banking of plasma (N~350-400 in FY20) – expect ~65% of participants to opt-in to donate 30 cc for plasma aliquoting (per BBDP processing); buffy coat sent to TGen for ApoE4-typing.

### **Proposed One-Year and Long-Term Outcomes:**

Clinical phenotyping of all participants, and N~350-400 plasma samples by end of year 1. NIH grant submission in 2021 on biopsychosocial determinants of cognitive resilience and reserve in the older old that utilizes a latent class analysis approach integrating manifest variables from demographics (age, education, gender), ApoE-status, clinical phenotype, blood biomarkers (e.g.  $\beta$ -amyloid and p-tau (AD), neurogranin (synaptic injury), NfL (axonal injury), and LCS psychosocial, cognitive and physical variables.

### **Year End Progress Summary:**

Since the beginning of the funding period that started on 7/1/2019, and through 2/29/2020 (the reporting period thus far), there has been great progress made and we are on schedule to fulfill the specific aims of the grant. We were able to get a running start on both aims by developing a protocol and submitting and obtaining IRB approval in order to begin implementation of clinical phenotyping classification (e.g. CDR) [Specific Aim 1] and blood draws [Specific Aim 2] in July and August 2019.

### **Progress made on Specific Aim 1 –**

We required that all new enrollees into the LCS have a study participant partner in order to undergo the CDR interview. We also asked previously enrolled participants who did not meet high thresholds on cognitive testing and trajectory (e.g. Montreal Cognitive Assessment, MoCA, scores of 27 or higher regardless of age and no decline in score of two or greater points compared to a previous score) to provide a study partner to undergo the CDR interview. Through February 2020, 300 of 371 (80.8%) eligible new or previously enrolled LCS participants were classified: 218 via CDR assessment (120 in LCS and 98 who are dually enrolled in LCS and BBDP) and 86 via

achieving high cognitive assessment test scores and identifying as being independent on activities of daily living. Seventy-one participants did not undergo CDR or in-person (MoCA) cognitive assessment: 33 declined or were not able to provide a study partner to undergo the CDR interview and 38 underwent telephone on in-residence abbreviated assessment only. As expected, the vast majority of active LCS participants (>96%) are thus far being classified as without dementia. The incidence of minimal or mild cognitive changes/impairments are thus far in the 25% range, which is within the expected range for participants who are, on average, in their 80's and of whom >40% are above age 84 years. We are also on track to recruit ~100 new participants in to the LCS in 12 months (62 newly enrolled in 8 months). This aim is on track to provide clinical phenotyping of LCS participants.

**Progress made on Specific Aim 2 –**

Response to requesting LCS participants to opt-in to donate ~30 cc of plasma for aliquoting, banking of plasma and sending the buffy coat to TGen for ApoE4-typing (per collaboration supported by a previous AARC grant to TGen) has been outstanding. We had aimed for ~65% of participants opting in to donate plasma, however, to the credit of the participants, 91.8% of eligible participants have donated a plasma sample – from July 2019 through February 2020, 323 samples were collected (225 LCS participants and 97 LCS and BBDP dually enrolled participants). Our goal had been ~350-400 samples to be collected in the 12-month grant period, and thus with 323 already collected in 8 months we expect to exceed this goal. ApoE4-typing is sent to TGen in batches of 86 to facilitate bulk processing – results on 187 are known (97 dually enrolled with BBDP and 90 from LCS only participants); 86 other samples were sent to TGen on 2/14/2020 with results pending; and the remainder are being held to be sent to TGen with the next batch sample.

In summary, there has been excellent progress made in the 8 months of funding (July 2019 through February 2020). We are not only on track to achieve the grants specific aims, but are highly likely to exceed the originally proposed goal (Aim 2) of collecting ~350-400 plasma samples. With completion of these aims we will be well-positioned to have foundational and necessary data for publications and submission of a grant application in FY2021.

**Project Progress Reports**  
**Barrow Neurological Institute**  
**At St. Joseph's Hospital and Medical Center**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Hispanic Enrollment in Alzheimer's Research Trials (the HEART Program at BNI).** Meredith Wicklund, MD, Anna D. Burke, MD. Barrow Neurological Institute; St. Joseph's Hospital and Medical Center; St. Joseph's Westgate Medical Center; Chandler Regional Medical Center; Mercy Gilbert Medical Center; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) Implementation of the HEART Program includes a formal development plan outlining internal and external outreach strategies to increase recruitment and the establishment of organizational infrastructure, resources, and written translational materials to promote trial retention while recognizing unmet needs of a large Spanish-speaking community seeking care within Maricopa County.
- 2) To forge a close working relationship with members of our Hispanic community to formalize the HEART outreach program to increase Alzheimer's disease awareness while addressing clinical research opportunities and family/caregiver support needs to increase trial retention through novel service-related solutions.
- 3) To identify and mitigate against cultural barriers limiting access for Hispanic patients to enroll into Alzheimer's disease clinical trials.

### **Background and Significance:**

Hispanics facing the problem of Alzheimer's disease (AD) constitute an underserved and understudied population in the United States. BNI has partnered with various organizations in the community to help address the educational and clinical needs of patients and families and to demonstrate to this underserved community our strong interest in understanding the unique factors affecting their cognitive health.

### **Proposed One-Year and Long-Term Outcomes:**

The HEART Program's outreach objective is designed around an internal (within BNI and Dignity Health opportunities) and an external outreach plan (community) for recruitment, with an established recruiter training program, metrics, and goals to maximize engagement among the Hispanic community. Our retention plan includes focused translational tools (such as Spanish translated rating scales) and expanded training among research team personnel offered by Promotores and Hispanic Community Stakeholders to address unique cultural needs. The HEART Program plans to recruit participants from the community through education, outreach, and various events such as memory screens. To support the core in recruiting, enrolling, and retaining 100 Hispanic participants, we will attend community events celebrating Hispanic culture, develop written materials, including a caregiver dementia handbook, in both English and Spanish to expand our reach, and partner with various agencies serving both English and Spanish-speaking Latino seniors. The enrollment goal for BNI will be to have 40-50 actively enrolled Hispanic participants by 2020.

### **Year End Progress Summary:**

- 1) We have recruited and trained Spanish speaking research personnel and raters to provide study visits in English or Spanish, at the participant's discretion.

- 2) We have partnered with the Promotores program to provide education to our research team on development of culturally sensitive education and outreach materials as well as foster collaboration in recruitment of Hispanic individuals.
- 3) We have developed a dementia caregiving guide with Spanish translation and materials sensitive to the Hispanic culture. The guide is currently undergoing final review and editing.
- 4) We have partnered with Latinos Against Alzheimer to provide education and outreach to the local Hispanic community, through co-sponsored events such as “Alzheimer’s and Other Dementias’ Impact in Communities of Color at Phoenix Brain Health Movie Night.”
- 5) We have partnered with the Center for Senior Living and other area senior organizations to provide free memory screenings, which has resulted in recruitment of several new participants.
- 6) We have partnered with area Hispanic media organizations to provide targeted education and outreach on Alzheimer’s disease the local Hispanic community. This has included appearances on local Spanish speaking radio stations and Facebook Live events for the Spanish speaking community.
- 7) We have recruited Spanish speaking Health Care Advisor and a Spanish speaking Social Worker to develop additional educational and outreach to the Hispanic community. They provide one on one counseling in the clinic for education, resources, behavioral management and other needs in caring for Hispanic individuals with dementia.
- 8) We have developed and implemented a monthly Memory Café for Spanish speaking individuals with dementia and their caregivers. The first event is in February 2020.
- 9) Through the above efforts, we have been able to recruit 2 new Hispanic participants since July 2019, with overall 17 active Hispanic participants. While this falls short of our one-year outcome, we have identified that financial barriers for caregivers to take time for work, provide transportation and support Hispanic participants in the AADC is a barrier. We have worked with the Arizona Alzheimer Consortium to provide stipends to offset the burden to participants. Additionally, the length of study visits has been identified as a barrier. We have worked with our collaborators to develop study materials with Spanish translation that provides culturally sensitive, requisite information while limiting study fatigue. We also note that recruitment has been short of goal because it takes time and effort to develop a meaningful, sustainable relationship with the Hispanic community. With addressing the barriers noted and the programs in development, we will be able to establish a firmer presence in the local Hispanic community to provide the education, outreach and recruitment services needed for this community and, as such, anticipate improved recruitment over the long-term. As evidence of this, we have already received referrals since January 2020 for 25 new Hispanic participants.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Collection of biofluids and generation of cell lines from AD and FTD patients.** Meredith Wicklund, MD, Anna Burke, MD. Barrow Neurological Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) Collect longitudinal clinical information, blood, CSF and urine samples from AD and FTD patients.
- 2) Process samples and store serum, plasma, CSF and urine samples in the Biobank at Dignity Health to make these samples available to the research community.
- 3) Isolate patient PBMCs for the generation of pluripotent stem cells.

### **Background and Significance:**

Alzheimer's disease (AD) and frontotemporal dementia (FTD) are neurodegenerative disorders that cause progressive cognitive decline in older adults. While a number of model systems and hypotheses regarding mechanisms regulating neurodegeneration during AD and FTD have been generated, a critical issue is the demonstration that these same mechanisms occur in the patient population. There remains an urgent need to collect biofluids (blood, CSF, urine) from AD and FTD patients for use in research studies. In addition, the generation of pluripotent stem cells from these patients will greatly facilitate research using patient derived cells. Such pluripotent stem cells can be generated using isolated PBMCs from patient blood samples.

### **Preliminary Data:**

We have significant experience within the BNI and Dignity Health to collect patient derived biofluids linked to clinical information, process and store biofluid samples for research purposes and the generation of adult stem cell lines. These activities are common in the neuromuscular disease clinic and the Biobank is currently processing and storing samples linked to clinical information for many projects, clinical research studies and trials throughout the hospital. We have recently initiated a web portal for investigators to search for samples within our Biobank that can be requested for research purposes. Therefore, the infrastructure is already in place to successfully complete the proposed study.

### **Experimental Designs and Methods:**

- 1) Collect longitudinal clinical information, blood, CSF and urine samples from AD and FTD patients.
- 2) Process samples and store serum, plasma, CSF and urine samples in the Biobank at Dignity Health to make these samples available to the research community.
- 3) Isolation of patient derived PBMCs.

### **Proposed One-Year and Long-Term Outcomes:**

We will initiate our biobanking efforts and collect biofluids and data from at least 25 subjects. Samples will be made available to investigators for research purposes using the Dignity Health Biobank web portal. We will seek external support to continue our biobanking efforts for AD and FTD. We also hope to enroll participants in post-mortem tissue banks so that we will ultimately have longitudinal biofluids and clinical information, patient derived stem cells, and post-mortem tissue samples. This provides an extremely valuable resource to the research community.

**Year End Progress Summary:**

- 1) In efforts towards developing a biobank, an IRB approved protocol has been established, allowing us to begin recruitment of participants.
- 2) Standard operating procedures (SOPs) have been developed for collection and storage of the biofluids in collaboration with laboratory personnel of the Dignity Health Biobank.
- 3) A databank has been designed and created for electronic storage of clinical information that will be linked to biofluids.
- 4) A protocol for sharing of clinical information and biofluids with the research community has been generated.
- 5) We have worked diligently to create and implement new SOPs for research and biobank staff. As this required development and validation of new protocols, needing final approval by multiple entities including IRB and biobank staff that took time to work through, there was unanticipated delay in initiating collection of biofluids from participants. As such, no collection of biofluids as of yet been collected but all SOPs and approvals are now in place. We have created and maintained a list of participants who have expressed interest in participation in research trials and agreed to be contacted for the same. These participants are being scheduled for study visits now. We do not anticipate further delays in recruiting the planned 25 participants.
- 6) We are aiming to create collaborations with the Barrow Neurological Foundation, Arizona Alzheimer Consortium and other partners for ongoing biobanking efforts to recruit participants beyond this study proposal. With continued funding, we aim to establish longitudinal collection of biofluids from participants and to coordinate with post-mortem

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Nucleocytoplasmic trafficking deficits of ADAR2 and RNA editing aberrations in Alzheimer's disease.** Rita Sattler, PhD, Kendall Van Keuren-Jensen, PhD, Elliott Mufson, PhD. Barrow Neurological Institute; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

Aim 1: Determine nuclear pore defects and ADAR2 nucleocytoplasmic mislocalization in postmortem tissue samples of different subgroups of AD (MCI, mild, severe).

Aim 2: Determine nuclear pore defects and ADAR2 nucleocytoplasmic mislocalization in AD patient-derived iPSC cortical neurons and test novel inhibitors of nuclear export.

Aim 3: Determine RNA editing changes using RNA sequencing technology in postmortem AD patient tissue samples and AD patient-derived iPSC cortical neurons.

### **Background and Significance:**

#### **Nucleocytoplasmic trafficking deficits**

Defects at the nuclear pore complex, which facilitates nucleocytoplasmic trafficking of mRNAs and/or proteins in and out of the nucleus, have been described by us and others as a critical component of disease pathogenesis in Amyotrophic Lateral Sclerosis (ALS)/Frontotemporal dementia (FTD), but has since also been detected in Huntington's disease (HD), Alzheimer's disease (AD) and tau-mediated FTD. The downstream consequences of these defects are still poorly understood. One hypothesis suggests that the mislocalization of RNA binding proteins, such as TDP-43, could be explained by these deficits. Our laboratory has now identified an additional RNA processing protein, ADAR2, to be mislocalized to the cytoplasm in ALS/FTD, and as a consequence initiates widespread RNA editing aberrations, which are suggested to be responsible for disease pathogenesis (*Moore et al 2019, Acta Neuropathologica*). Similar mechanisms are thought to play a role in AD.

#### **RNA editing**

RNA editing is a crucial component of the cellular RNA homeostasis ranging from alterations of the amino acid sequences, alternative splicing, gene expression and miRNA binding. Therefore, it is not surprising that dysregulation of the RNA editing machinery has been associated with the pathogenesis of numerous neurodegenerative and neurological disorders.

### **Preliminary Data, Experimental Design and Methods:**

#### **ADAR2 mislocalization in ALS/FTD**

Our laboratory has shown recently that ADAR2 is mislocalized to the cytoplasm in C9orf72 ALS/FTD postmortem patient tissue, patient-derived iPSC motor neurons and a C9 mouse model.

#### **ADAR2 mislocalization in AD**

Similar to what we showed in ALS patient tissue, we have preliminary data showing ADAR2 mislocalization in the frontal cortex of AD patient tissue. While not significant in mild cognitive impaired or mild AD patients, we do see significant mislocalization in severe AD patient tissue.

#### **RNA editing aberrations due to ADAR2 mislocalization in C9orf72 ALS/FTD**

Comparison of A to I editing ratios in C9orf72 ALS/FTD tissues yielded 11,466 aberrantly editing RNA editing sites spanning 1,458 genes (data not shown). Unsupervised hierarchical clustering on RNA editing analysis resulted in the segregation of C9orf72 ALS/FTD spinal cord and non-ALS control spinal cord tissue. Gene Ontology Analysis of all genes exhibiting RNA editing aberrations revealed that transcripts related to ALS and the EIF2 signaling pathway are the most

significantly mis-regulated pathways in C9orf72 ALS/FTD (data not shown). These data suggest that similar to ALS, RNA editing changes in AD may play a significant role in important disease pathways, which could be therapeutically targeted with compounds targeting the nuclear pore complex, such as KPT compounds.

### **Experimental Design and Methods:**

#### **Specific Aims 1+2**

##### **Immunohistochemistry and immunofluorescent staining of AD patient postmortem tissue**

Standard immunohistochemistry and immunofluorescent protocols will be applied to stain patient tissue for nuclear pore proteins as well as RNA binding proteins. We will monitor protein localization of the following proteins: ADAR2 (anti-ADAR2, Sigma-HPA018277), TDP-43 (anti-TDP-43, Abnova-H00023435-M01), lamin-B (anti-lamin-B1, Santa Cruz- sc-374015), Nup414 (anti-Nup414, BioLegend- MMS-120P). To ensure nuclear neuronal localization, tissue and cells will be co-labeled for neuronal marker protein MAP2 (anti-MAP2, Synaptic Systems-188 009) and nuclear marker DAPI (Life Technologies-P36930).

Tissues will be obtained from Dr. Mufson's tissue collection, and if additional tissue is needed, from the Brain and Body Donation Program at Banner Sun Health Research Institute. We will focus our analyses on frontal cortex regions.

##### **Immunofluorescent staining of AD patient iPSC cortical neurons**

AD patient iPSCs (sporadic, presenilin, APOE4) will be differentiated into cortical neurons using existing and established protocols. Similar to the patient tissue, iPSC-CNs (at 77 days after differentiation) will be examined for protein localization of nuclear pore membrane proteins lamin-B and Nup414, as well as ADAR2 and TDP-43, in the presence of neuronal marker MAP2 and nuclear marker DAPI. If we find ADAR2 mislocalization, we will treat cells with selective inhibitors of nuclear export (KPT compounds), which are currently being moved into clinical trial for ALS [6, 11].

Patient iPSC lines have already been obtained by Dr. Dave Brafman, ASU.

#### **Specific Aim 3**

##### **RNA sequencing and RNA editing analysis of patient tissue and iPSC-CNs**

RNA will be isolated using RNAqueos – Micro Total RNA Isolation kit and sequencing libraries will be prepared using SMARTer Stranded Total RNA-Seq Kit v2 – Pico Input (Takara Bio – 634413). Libraries will be combined into equimolar pools and sequenced on an Illumina paired-end flowcell (Illumina – PE-401-3001) with a 1% v/v Phix v3 spike-in (Illumina – FC-110-3001) on Illumina's HiSeq 2500 with TruSeq v3 chemistry (Illumina – FC-401-3002). Next we will perform RNA A to I editing analysis at 408,580 known RNA editing sites, a method now commonly used in our laboratory.

### **Proposed One-Year and Long-Term Outcomes:**

This proposal will provide sufficient data to determine whether nucleocytoplasmic trafficking defects in AD lead to ADAR2 mislocalization and RNA editing aberrations. Follow up studies will allow us to study in more detail how the RNA editing changes of specific genes/pathways will impact neuronal function and survival. With the pilot data obtained from this proposal we will be able to apply for federal funds (NIH) to support these follow up studies.

### **Year End Progress Summary:**

We have been able to select AD patient tissue through the collaborative support from Dr. Elliott Mufson. In addition, we have propagated and quality controlled (pluripotency markers, proper karyotyping, genetic mutations) and frozen down the AD lines obtained from Dr. Dave Brafman.

While accomplishing these goals, and prepping for immunohistochemistry experiments, we were notified by our ADAR2 antibody supplier (Sigma/Atlas HPA018277) that this antibody was discontinued.

We then tested 4 different antibody sources (6 different antibodies in total) for ADAR2 antibodies, and tested those on human tissue as well as human iPSC neurons – but were unable to select an antibody with enough stringent specificity required for our analyses.

Antibody testing experiment:

Please see below the progress we made to overcome these difficulties:

1. We since entered a collaborative agreement with the original antibody generating company (Sigma via Atlas Antibodies AB) to re-generate their ADAR2 antibody. This project is ongoing and as soon as we receive and have validated the newly produced antibody, we will continue our proposed studies for ADAR2 mislocalization in AD. Once mislocalization is confirmed, we will then pursue the RNA sequencing experiments to determine the proposed RNA editing aberrations.
2. To be able to follow ADAR2 mislocalization in iPSC cortical neurons, we developed a lentiviral construct overexpressing ADAR2-GFP. This construct is currently being validated in immortal mammalian cell lines and healthy control iPSC cortical neurons. Once validated, we will use this overexpression virus to treat AD iPSC cortical neurons and examine ADAR2-GFP localization over time.
3. We are working with Dr. Patrick Pirrotte (TGen) to develop a Mass Spec approach to detect ADAR2 peptides which we can then apply to quantitative detection of ADAR2 in iPSC cortical neuron cytoplasmic fractions.
4. Finally, in collaboration with Dr. Daniela Zarnescu (UofT) we are growing and expanding a *Drosophila* ADAR-HA fly strain, which will allow us to cross these flies to any fly AD models that are available in the fly community and then follow ADAR-HA localization in a disease background.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Development of a Multi-Scale MRI Method for Preclinical Validation of Hemodynamic Factors Associated with AD.** Ashley M. Stokes, PhD, Maurizio Bergamino, PhD, Salvatore Oddo, PhD. Barrow Neurological Institute; Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

**Aim 1: Develop and optimize the SAGE perfusion MRI method on the Bruker preclinical system**

We will develop our advanced multi-scale, multi-parametric SAGE DSC-MRI method on the preclinical imaging system in order to characterize complementary hemodynamic metrics, including macro- and microvascular perfusion, vascular architecture, and blood-brain barrier breakdown. This method will be compared to the currently available method (arterial spin labeling (ASL)) in the 3xTg mouse model. We hypothesize that SAGE-perfusion will provide more robust and comprehensive perfusion estimates than ASL perfusion.

**Aim 2: Assess vascular and metabolic function in the 3xTg mouse model of AD**

We will assess complementary vascular and metabolic biomarkers using SAGE-perfusion MRI and FDG-PET imaging, respectively, in the 3xTg mouse models and wild-type mice (WT). We will correlate regional hemodynamic metrics with FDG-PET metrics. We hypothesize that microvascular hemodynamic metrics and BBB disruption will be significantly altered in 3xTg vs. WT, and that regional microvascular changes will co-localize with reduced metabolic activity.

### **Background and Significance:**

Methods to assess novel vascular metrics may provide new insight into the association between vascular risk and AD. Recent studies have found decreased microvascular density in a transgenic amyloid mouse model compared to control mice (15), and other mouse models of AD have shown significant morphological and architectural changes in the vasculature (16). This study aims to further characterize vascular and metabolic changes in preclinical models of AD using advanced MRI metrics, plus standard FDG-PET. This will allow us to investigate multi-scale perfusion and metabolism as complementary biomarkers of AD-related neurovascular changes, leading to a much broader understanding of the impact of neurovascular dysfunction in AD.

### **Preliminary Data**

We previously acquired SAGE-perfusion data in the C6 rat brain tumor model. Maps of micro and macrovascular cerebral blood flow (CBF) and volume (CBV) demonstrate elevated perfusion associated with the tumor relative to contralateral normal tissue. We have also applied this method to rat models of stroke (11). Using the 3xTg mouse model (in collaboration with Dr. S. Oddo, Co-I), we previously assessed regional neurodegeneration and ASL-based perfusion compared to the 3xTg-S6K1 and WT mice. We found that genetic reduction of S6K1 led to salvaged cortical volume and blood flow, while hippocampal volume was not impacted.

### **Experimental Designs and Methods**

We have significant expertise in development of advanced MRI pulse sequences. The SAGE method will be developed using EPI readouts within a currently available single-line version of SAGE (programmed by AMS, PI) on the Bruker 7T MRI. Subsequent data will be acquired at a spatial resolution of 0.56 mm in-plane and 1 mm through-plane, with partial Fourier encoding to achieve TEs between 8ms and 100ms, and a temporal resolution of 1s. We will acquire SAGE-MRI and FDG-PET imaging in 20 mice (10 3xTg and 10 WT). For perfusion, SAGE-MRI images

are acquired during injection of 0.2 mmol/kg Gd-based contrast agent. Post-processing will be performed using standard methods (17). Mice will undergo FDG-PET within 24 hours to assess metabolic activity using standardized uptake value ratio (SUVR) maps.

### **Proposed One-Year and Long-Term Outcomes:**

This funding will allow us to assess vascular and metabolic features in the 3xTg mouse model. We anticipate publication of these results, and we plan to leverage this data in an NIH/NIA application to validate the underlying association between vascular and metabolic biomarkers in AD mouse models, in combination with histology and amyloid and tau PET.

### **Year End Progress Summary:**

**Aim 1:** The goal of Aim 1 is to develop and optimize the SAGE perfusion method for the Bruker 7T preclinical scanner. We implemented the SAGE method on Bruker software with flexibility for various pulse sequence parameters (including voxel geometry, repetition time, echo times and echo spacing, number of echoes both pre- and post-refocusing pulse, and readout time). The resulting sequence is highly flexible. This method was compiled and installed on the Bruker 7T MRI; the method was subsequently tested in both rats and mice, with and without an injection of contrast agent. The optimized set of parameters included 4 echoes total, with 2 gradient-echoes (echo times 5.73 and 14.2 ms), 1 asymmetric spin-echo (echo time 32.4 ms), 1 Hahn spin-echo (echo time 40.8 ms). Voxel geometry was 0.25 x 0.5 mm<sup>2</sup> in-plane resolution, 2 mm slice thickness, with 4 coronal slices. Other parameters include 570 ms repetition time with 635 dynamic scans. The total scan time for perfusion imaging is 6 minutes, which should enable the quantification of both perfusion and permeability.

**Aim 2:** We obtained 28 mice from Dr. Salvatore Oddo (ASU, Co-I), of which 15 were 3xTg-AD and 13 were WT. All mice were female, in accordance with the model pathology. At the time of scanning, mice were 15 months of age (15.06 (standard deviation 0.10) months for 3xTg, 15.08 (SD 0.07) months for WT). Four of the 3xTg mice and one of the WT mice died before or during scanning. As of 3/1/2020, all 3xTg scans have been completed, and 5 of the 12 remaining WT mice have been scanned. The remaining scans are scheduled to occur the first week of March. All mice underwent surgery the day before scanning to insert jugular catheters. MRI data (Bruker Biospec, 7T) included T2-weighted images, 3D anatomical scans for parcellation, diffusion tensor imaging (DTI) to assess microstructural integrity, pre-contrast T1-weighted images, and pre-contrast T1 maps. During dynamic SAGE imaging, 0.1 mmol/kg of gadolinium-based contrast agent (Gadavist) was injected using a power injector via jugular catheter. Perfusion data was acquired for 6 minutes, with 30 seconds of baseline. Following perfusion imaging, post-contrast T1-weighted images and T1 maps were acquired. The total MRI protocol was 75 minutes. For FDG-PET, mice were fasted for four hours prior to scanning. Immediately before injection of FDG, blood glucose was measured using a standard blood glucometer. Mice were injected with approximately 0.54 mCi of FDG. PET images (Bruker Albira) were acquired from 40 to 70 minutes after injection to assess glucose metabolism. On the following day, all mice were sacrificed and brains extracted for genetic and histological analysis.

Preliminary data analysis showed excellent image quality, and advanced metrics within expected physiological limits. A multi-atlas label fusion method has been developed to assess volumetric differences between the groups, including the whole brain, hippocampus, cortex, total ventricles, and caudate putamen. Total and microvascular cerebral blood volume (CBV) are quantified using an advanced perfusion imaging method based on SAGE contrast. DTI data are processed using DSI Studio to quantify microstructural white matter changes, including fractional anisotropy, mean diffusivity, and axial and radial diffusivities. Glucose metabolism will be compared using ROI analysis of standardized uptake value ratio (SUVR) maps. Data analysis is ongoing.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Multi-Scale MRI Assessment of Neurovascular Factors Associated with AD.** Ashley M. Stokes, PhD, Maurizio Bergamino, PhD, Yi Su, PhD, Leslie C. Baxter, PhD, Anna Burke, MD  
Barrow Neurological Institute, Banner Alzheimer's Institute, Mayo Clinic Arizona, and Arizona Alzheimer's Consortium.

### **Specific Aims:**

#### **Aim 1: Establish micro- and macro-vascular hemodynamic signatures along the AD spectrum**

We will implement our advanced multi-scale, multi-parametric SAGE DSC-MRI method to characterize complementary hemodynamic metrics, including macro- and microvascular perfusion, vascular architecture, and blood-brain barrier breakdown, in older subjects with and without cognitive impairment. We will correlate regional hemodynamic metrics with cognitive metrics and amyloid-PET metrics. We hypothesize that microvascular hemodynamic metrics and BBB disruption will be significantly altered in patients with cognitive impairment. Regional microvascular changes identified using this method will co-localize with elevated amyloid burden.

#### **Aim 2: Establish micro- and macro-vascular functional activation patterns along the AD spectrum**

We will compare multi-scale SAGE fMRI response and activation patterns in the same subjects from Aim 1 using task-based fMRI paradigms (including episodic memory and self-reflection tasks). Using whole-brain analysis, we will correlate micro- and macrovascular response with clinical cognitive metrics and amyloid-PET metrics. We hypothesize that micro- and macro-vascular SAGE-fMRI mismatch will correlate with domain-specific neurocognitive deficits and PET amyloid burden in patients with cognitive impairment.

### **Background and Significance:**

Vascular changes are known to occur prior to cognitive decline and may be detectable earlier than standard biomarkers. This has led to the development of advanced MRI methods that are sensitive to neurovascular characteristics: perfusion to cerebral blood flow and hemodynamics, permeability to blood-brain barrier degeneration, and fMRI to synchronous fluctuations in brain activity. While substantial evidence implicates AD-related neurovascular changes, one limitation is that a comprehensive assessment of neurovascular factors is lacking with current methods. To overcome this limitation, we propose to investigate multi-scale perfusion and functional activation as complementary biomarkers of AD-related neurovascular changes, leading to a much broader understanding of the impact of neurovascular dysfunction in AD.

### **Preliminary Data:**

We previously acquired SAGE-DSC data in a 67-year-old female with a history of metastatic breast cancer, who presented with memory loss and weakness. In this subject, the hippocampus demonstrated higher macrovascular CBV and CBF than cingulate cortex, but lower microvascular CBF; in contrast, hippocampal macrovascular CBF was elevated relative to thalamus, while microvascular CBF was reduced.

SAGE-fMRI was tested in a healthy young volunteer using a vision task, with comparison to standard fMRI. SAGE  $R_2^*$  activation closely matches that of the standard fMRI activation but is less sensitive to anomalous clusters (blue arrows). SAGE  $R_2$  activation is more localized to microvascular activation and less sensitive to large draining vessels. While the  $R_2$  activation appears attenuated, it likely reflects more closely underlying microvascular activation.

### **Experimental Designs and Methods:**

We will recruit a total of 6 people (3 each with and without cognitive impairment). All subjects will undergo a cognitive battery immediately prior to MRI. MRI data acquisition will be performed at BNI using a dedicated research 3T Philips MRI. The MRI protocol will include standard structural images, SAGE-fMRI, and SAGE-perfusion imaging. For SAGE-fMRI, multi-echo SAGE images are acquired during memory-related task paradigms. For perfusion, SAGE-MRI images are during injection of 0.1 mmol/kg Gd-based contrast agent. Post-processing will be performed using standard methods. Within 2 weeks of MRI, patients will undergo PET imaging using <sup>18</sup>F-Florbetaben at Banner Alzheimer's Institute. The PET scan will be performed on a GE Discovery PET/CT scanner after a bolus injection of approximately 8 mCi of NeuraCeq. A standard PET analysis pipeline will be used to generate standardized uptake value ratio (SUVR) maps, regional SUVR, and overall amyloid burden index. Determination of amyloid positivity will be based on published SUVR cutoffs.

### **Proposed One-Year and Long-Term Outcomes:**

Given the budgetary constraints, this funding will allow us to generate pilot data using both PET and MRI in a few subjects. We plan to leverage this data in our NIH/NIA application, which will support PET and MRI data, along with genetic and clinical biomarkers, in a larger subject cohort.

### **Year End Progress Summary:**

Two subject groups were recruited for this study: cognitively normal (CN) (n = 5) and cognitively impaired (CI) (MCI and mild to moderate AD) (n = 4). All subjects were recruited for the perfusion and functional MRI, and a subset of 4 subjects were simultaneously enrolled for the amyloid PET scans.

Subjects underwent cognitive testing using the Montreal Cognitive Assessment (MoCA) and Hopkins Verbal Learning Test (HVLT), along with other cognitive tests prior to MRI. MRI data were acquired at 3T (Ingenia, Philips) at the Barrow Neurological Institute. Structural MRI data was obtained using ADNI (Alzheimer's Disease Neuroimaging Initiative) protocols. Diffusion tensor imaging was acquired to assess microstructural integrity. Advanced MRI methods were used to measure functional activation (via the fMRI blood oxygen level dependent (BOLD) response) and perfusion metrics cerebral blood flow (CBF) and volume (CBV) (via perfusion MRI acquisition during injection of Gd-based contrast agent). Perfusion MRI and fMRI were acquired using an advanced multi-echo, multi-contrast MRI technique that enables separation and quantification of total and microvascular characteristics. Tasks include vision, face-name encoding and retrieval, and a self-reflection task. Perfusion data were acquired during injection of 0.1 mmol/kg gadolinium-based contrast agent. Within one month of MRI, a subset of subjects (n = 4) underwent amyloid PET scanning with <sup>18</sup>F-Florbetapir at Banner Alzheimer's Institute.

Preliminary trends between CN and CI groups are indicative of hypoperfusion in the CI group. These results could be indicative of varying microstructural traits between groups. The control fMRI task (vision) reveals no apparent difference in BOLD response between CN and CI groups, as expected. Analysis of the memory-associated fMRI tasks is ongoing.

Based on the preliminary data, we anticipate submission of a new R01 application (PI: AM Stokes) entitled *Multi-Scale MRI Assessment of Neurovascular Factors Associated with AD*. This proposal will be submitted to NIA in June 2020. We are currently preparing a publication for the functional MRI analysis methods (demonstrated in healthy volunteers), which should aid this application. In addition, we have been in discussions with a company (Cercare Medical, Denmark) that develops software for perfusion analysis specifically targeted at aging cohorts. This application will leverage these recent developments and will be further aided by the data acquired in this study.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Splicing and tau pathology in Down syndrome and Alzheimer's disease.** Sylvia E Perez, PhD, Elliot J Mufson, PhD, Rita Sattler, PhD, Bin He, MD, Ileana Lorenzini, PhD. Barrow Neurological Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

Individuals with Down syndrome (DS), a genetic disorder caused by an extra copy of the human chromosome 21 (HSA21), have by the fourth through sixth decade of life an increased risk to develop Alzheimer's disease (AD) type-dementia (D) associated with tau-containing neurofibrillary tangles (NFTs) and  $\beta$ -amyloid (A $\beta$ ) plaque pathology<sup>1,2,3</sup>. DS population has almost complete penetrance of AD in contrast to the normal disomic population which rises with age. Interestingly, only about two thirds of DS cases develop dementia<sup>4,5</sup>, making DS an excellent natural genetic clinical pathologic model to study AD-like cellular and molecular mechanisms that underlie dementia. We recently reported that people with DS with dementia (DS+D) display more advanced NFT tau pathology in frontal cortex (FC) layer V and VI projection neurons, compared to those without dementia (DS-D)<sup>6</sup>. Although these FC NFT positive neurons contain a different genetic signature compared to DS-D, a similar FC  $\beta$ -amyloid (A $\beta$ ) plaque load was found between DS groups<sup>6</sup>, suggesting that tau pathology plays a key role in dementia onset in DS. However, there is virtually no information identifying the molecular/cellular mechanism that underlie the differential tau pathobiology seen in FC neurons between DS+D and DS-D. Recently, Bai and colleagues<sup>7</sup> demonstrated a mislocalization of the splicing U1-70k and U1A small nuclear ribonucleoproteins (snRNP) in NFT-like structures in AD and DS<sup>8</sup>, revealing a disruption of mRNA processing, that may play a critical role in the NFT pathogenesis in AD and DS. However, the mechanism involved in U1-70K and U1A dysregulation and its relationship with other RNA splicing proteins and tau epitope pathologic progression within FC projection neurons in DS+D, DS-D and AD remain unknown. Remarkably, we found an increase in NFT-like structures positive for U1-70K, a reduction in nuclear area for the nuclear splicing factors SR splicing factor 2 (SRSF2 or SC35) and SR repetitive matrix protein 2 (SRRM2) in FC projection neurons in DS+D and diminution in heterogeneous nuclear (hn) RNP A2/B1 in DS (**Preliminary Data**). In addition, extranuclear and nuclear U1-70K positive neurons colocalized/coexisted with phosphorylated (AT8) and apoptotic (TauC3) tau markers in DS (**Preliminary Data**). Pathological imbalance between 3R/4R tau isoforms<sup>9-11</sup>, drives the formation of NFT and ultimately neuronal death<sup>12</sup>, which are pathologic hallmarks of DS and AD and correlate with severity and cognitive decline in AD and DS<sup>13,14</sup>. Interestingly, alterations in splicing proteins change the ratio of 3R to 4R or 4R to 3R<sup>11,15</sup>. Together these findings suggest that spliceosome dysfunction disrupts tau processing in DS. This proposal we will test the **hypothesis that splicing dysfunction is an early event in NFT formation in DS**. To define the cellular and molecular factors underlying spliceosome alterations and tau in cortical neurons in DS+D, DS-D and AD, we will use a novel approach of combining splicing and tau immuno-labeling with single-cell gene array technology. We will also use DS human Induced Pluripotent Stem cells (hiPSC) Cortical Neurons (CNs) to determine mechanistic interactions of these proteins<sup>16</sup>. The findings generated from these studies are crucial for development of novel therapeutic interventions to slow the onset of cognitive decline in DS and AD and aid in the advancement of novel biomarkers for these disorders. We will test the following interrelated specific Aims:

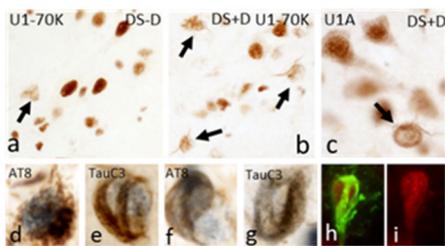
**Aim 1:** We will test the hypothesis that FC pyramidal neurons bearing phosphorylated (AT8) and truncated (TauC3) tau epitopes display an increase in extranuclear U1-70K and U1A concomitant

with a decrease in SRRM2, SRSF2 and hnRNP A2B1 splicing proteins in DS+D compared to DS-D, respectively, using quantitative immunohistochemical techniques.

**Aim 2:** We will test the hypothesis FC neurons displaying containing greater extranuclear U1-70K and U1A and a reduction in SRRM2, SRSF2 and hnRNP A2B1 splicing proteins associate with early and late tau epitopes display differences in classes of genes related to apoptosis and downregulation of tau phosphorylation, spliceosomal RNAs, in DS+D compared DS-D using quantitative immunohistochemical techniques combined with single cell transcript expression.

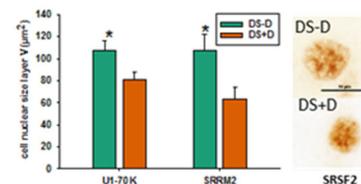
**Aim3:** We will test the hypothesis that induced-spliceosome protein dysregulation lead to an imbalance of 3R/4R tau transcripts in DS hiPSC-CNs.

**Background and Significance:** Although on about 2/3 of individuals with DS develop dementia, virtually all display amyloid plaques and tau tangles at forty years of age<sup>1-5</sup>. Over the last many years research has concentrated on the role the amyloid deposition play in DS+D cases. With the lack of efficacy of anti-amyloid vaccinations to meet their cognitive endpoints, both the AD and DS fields are attacking the role of tau microtubule-associated proteins involved in cytoskeleton functions including the role of the ratio of tau isoforms to the formation of NFTs<sup>17</sup>. Adult human brain expresses six isoforms of tau as resulted of constitutive and alternative splicing of the tau transcript<sup>18</sup>. The imbalance between 3R/4R tau isoforms, which in normal conditions is approximately 1, has been observed in several tauopathies clinically characterized by dementia, including AD and DS<sup>18</sup>. In fact, the disruption of the alternative splicing of tau exon 10 causes

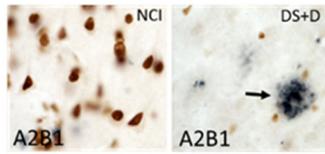


**Figure 1.** a-b, Images showing more cytoplasmic U1-70K positive (arrows) NFT-like structures in the FC layer V of DS+D compared to DS-D. c, Image showing extranuclear staining for U1A (arrow). d-i, Nuclear and extranuclear U1-70K (blue, red) colocalize with AT8 (brown: d and f; green: h) and TauC3 (brown, e and g) in both DS+D (d, e, h, i) and DS-D (f, g).

altered expression of 3R/4R tau isoforms leading NFT formation<sup>18</sup>, suggesting a critical role of splicing process in dementia onset. Splicing is a highly regulated process consist of intronic sequence elimination within a precursor mRNA to generate a mature mRNA. Constitutive and alternative splicing takes place in a dynamic nuclear ribonucleoprotein complex termed the spliceosome, comprised of five small nuclear (sn) RNAs (U1, U2, U4, U5 and U6), snRNP, SR and hnRNP proteins<sup>19</sup>. Recent studies, demonstrated U1-70K and U1A, displayed a widespread aberrant extranuclear accumulation in form of NFT-like structure in AD compared to preclinical AD stages suggesting that neuronal mRNA processing are compromised early in the disease and may play a key role in AD pathogenesis<sup>8</sup>. Remarkable, here we show that extranuclear mislocalization of U1-70K and U1-A occurs in FC of projection neurons of DS+D compared to DS-D (**Fig. 1**), indicative of early splicing dysfunction in DS. Likewise, nuclear and extranuclear U1-70K co-localize with the early phosphorylated (pSer<sup>202</sup>/The<sup>205</sup>) AT8 and late truncated (Asp<sup>421</sup>) TauC3 tau markers in DS FC neurons, suggest the involvement of U1-70K dysregulation in NFT pathogenesis in DS (**Fig. 1**). Besides snRNPs, SR proteins are also crucial for alternative splicing, which provides one of the most powerful ways of gene expression regulation by selecting a combination of exons, leading to the generation of many isoforms from a common transcript, as is in the case of the tau transcript<sup>20</sup>. In particular, alterations in the SRSF2, which is involved in the inclusion of the exon 10 (4R tau isoform), resulted in dysregulation of exon 10 and an imbalance of tau isoforms<sup>21,22</sup>. We have observed a reduction in nuclear size of the SRSF2 and SRRM2 in the FC neurons of DS+D compared to DS-D (**Fig. 2**), revealing also a dysregulation of alternative splicing that might lead to an imbalance of tau transcripts in demented DS cases. Likewise, hnRNP A2/B1 was



**Figure 2.** Shows a significant reduction in nuclear size for U1-70K, SRSF2 and SRRM2 splicing markers in DS+D compared to DS. \*p<0.05.

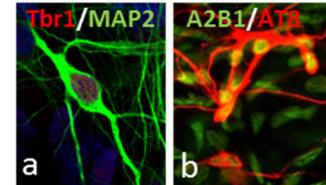


**Figure 3.** Images showing a substantial reduction of FC nuclear A2B1 staining (brown) in NCI compared to DS+D. Arrow indicates a plaque (blue).

downregulated in the FC neurons in both DS groups (**Fig. 3**). hnRNP A/B are splicing proteins involved in mRNA stability, maturation and trafficking<sup>23</sup> and are dysregulated in AD<sup>24</sup> and FTL/ALS<sup>25</sup>. These preliminary observations suggest that spliceosome dysregulation is broader and more advanced in DS+D and are key to dementia onset. Whether these alterations in spliceosome proteins could be prompted by APP/A $\beta$  favoring 3R or 4R tau isoforms in DS will be explored here using hiPSC-CNs (**Fig. 4**). Data generated will provide pivotal findings necessary to generate new drug targets for DS and AD.

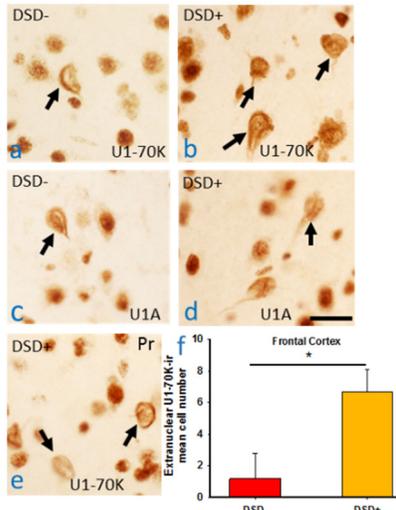
### **Preliminary Data, Experimental Design and Methods:**

**Methods and Experimental design.** Brain tissue has been and continues to be provided by the members of the national and international DS BioBank Consortium (DSBC) founded by Drs. Granholm, Head and Mufson and supported by the Bright Focus Foundation. We will use 4% paraformaldehyde-fixed tissue from 6 adult non-demented and 11 demented DS cases, 10 NCI and 10 AD cases. DS cases with and without dementia were obtained from members of the DSBC including UCI-Alzheimer Disease Researcher Center and Barrow Neurological Institute<sup>6</sup>. NCI and sporadic AD tissue collected from previous Rush ROS cases that are part of Dr. Mufson's PPG grant are already in-house. Human iPSCs from 4 trisomic DS and 1 disomic cell line (in-house) will be differentiated into CNs in collaboration with Dr. Sattler at the BNI. **Aim 1.** We will test the hypothesis that FC pyramidal neurons bearing phosphorylated (AT8) and truncated (TauC3) tau epitopes display a gradient of an upregulation of miss-located nuclear U1-70K and U1A concomitant with a decrease in SRRM2, SRSF2 and hnRNP A2B1 splicing proteins in DS+D compared to DS-D, respectively. To determine the relative numbers and expression of splicing markers during the evolution of tau pathology, FC sections will be doubly immunostained with either, U1-70K, U1A, SRRM2, SRSF2 and A2B1 and the AT8 or TauC3 tau epitopes in layers III and V, using an unbiased stereology procedure and densitometry<sup>26, 27</sup>. For data analysis we will use one-way ANOVA or Kruskal-Wallis test, and correlations with a Pearson or a Spearman rank test. Statistical significance will be set at 0.05. Immunohistochemical controls include peptide pre-adsorption and primary antibody deletion. **Aim 2.** We will test the hypothesis FC NFTs containing greater extranuclear U1-70K and U1A and a greater reduction in SRRM2, SRSF2 and hnRNP A2B1 splicing proteins associate with early and late tau epitopes display an upregulation of classes of genes related to apoptosis, downregulation of tau phosphorylation, spliceosome RNAs in DS+D vs DS-D. Single population expression profiling using custom-designed microarray analysis will be evaluated using the same cases from Aim 1. Fixed sections will be double immunostained using either U1-70K, U1A and AT8 and TauC3 tau markers. Double (nuclear and/or extranuclear U1-70K+AT8; U1-70K+TauC3; U1A+AT8, U1A+TauC3) and single (nuclear U1-70K and U1A) labeled cells from layers III and V will be micro-aspirated using a Zeiss laser capture microscope (LCM), mRNA will be extracted and custom-designed microarrays<sup>6,28, 29</sup>. The current array platform includes ~860 genes relevant to neurodegeneration, cell survival, cell proliferation, apoptosis, epigenetics, and RNA splicing. Approximately 50 individual labeled cells will be micro-dissected via LCM and captured per reaction for custom-designed array analysis<sup>6</sup>. Experiments are run in triplicate and blinded. Single cell RNA profiling is validated using qPCR. Relative changes in individual mRNAs will be analyzed by one-way ANOVA and adjusted using a false discovery rate (FDR) to reduce Type I error<sup>30</sup>. P value is set at 0.05. **Aim 3.** We will test the hypothesis induced-spliceosome lead to an imbalance of 3R/4R tau transcripts and increase in tau phosphorylated species in DS hiPSC-CNs. hiPSCs will be differentiated into CNs determinate by



**Figure 4.** hiPSC-CNs showing the expression of the cortical marker *Tbr1* (a, red), neuronal marker *MAP2* (a, green), *A2B1* (b, green) and phosphorylated tau marker, *AT8* (b, red).

the presence of cortical progenitor markers Brn2 (I-IV) and Tbr1 (V-VI)<sup>31-33</sup>. At day 75 of differentiation, cell lines are treated with fibrillary  $\beta$ -amyloid<sub>42</sub> (A $\beta$ <sub>42</sub>) and phosphorylation SR kinase inhibitors, lithium chloride (GSK-3 $\beta$ ) and AR-A014418 (Clk1). After three days of treatment cells will be fixed in 4% paraformaldehyde for 20 min or pelleted for RNA isolation. Counts and relative expression of the splicing proteins and tau phosphorylation markers will be done combining immunofluorescence and densitometry. Transcript expression will be evaluated using a custom-designed gene array using Nanostring technology. Gene dysregulation will be validated with immunofluorescence, western blot or qPCR. Data analysis we will use one-way ANOVA or Kruskal-Wallis test, Pearson or a Spearman rank test, and p-values (0.05) will be adjusted using a FDR to reduce Type I error.



*Figure 1. The cytoplasmic mislocalized U1-70k-ir NFTs are significantly increased in the FC layer V in DSD+. FC images showing many more U1-70k and U1A positive NFT-like structures (black arrows) in FC layer V of DSD+ compared to DSD-; positive U1-70k NFT- structures were also observed in layer V of Pr. U1-70k NFTs were significantly higher in DSD compared to DSD-.*

array. Respect **Aim 3**, hiPSCs differentiated into cortical neurons were treated with fibrillary  $\beta$ -amyloid (A $\beta$ ) and kinase inhibitors, lithium chloride (GSK-3 $\beta$ ) and AR-A014418, and analyses is ongoing. Below we presented the results obtained by challenging the hiPSCs-CN with fibrillary beta amyloid.

**Aim 1:** We found a significant increase in the number of FC projection neurons bearing mislocalized U1-70K and U1A in layer V in DSD+ compared to DSD- (**Figure 1**). Mislocalized U1-70K and U1A in layer V were also seen in the precuneus of DS (**Figure. 1e**). We showed a significant positive correlation between the numbers of FC projection neurons bearing extranuclear U1-70K-NFT like structures and the number of FC AT8 positive NFTs in layer V-VI (**Figure. 2**), but not with TauC3 in DS, linking U1-70K and U1A dysregulation to NFT pathogenesis. These findings reveal splicing loss/dysfunction related to tau pathology at the core of

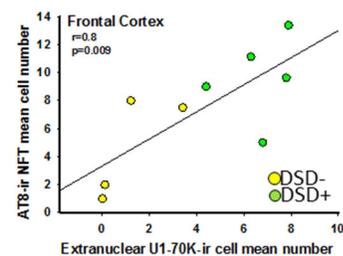
### **Proposed One-Year and Long-Term Outcomes:**

**Plan for external funding:** Preliminary will support federal (R21) and private (Alzheimer Association) grants.

**Future Studies:** To further unravel the molecular and cellular mechanisms involved in splicing dysregulation and the evolution of tau pathology in DS, we will characterize the nucleocytoplasmic transport and nuclear envelope structure, which are key players in alternative and constitutive splicing, and have been shown to be disrupted by pathological tau in AD<sup>34</sup>.

### **Year End Progress Summary:**

In the past six months we have accomplished some of the goals described in the **Aim 1**; however, **the Aim 2**, due to software complications that made the LCM unusable, a new core LCM was bought, which arrived and was installed two weeks ago, delaying the single cell gene-array assay in DS. Fortunately, we received **frontal cortex (FC)** and **precuneus (Pr)** frozen samples from demented and non-demented DS cases, allowing us to generate a general gene-array profiling in these two cortical areas using a custom-designed gene array with Nanostring technology in substitution of the single-cell



*Figure 2. shows a positive correlation between the numbers of AT8-ir NFTs and extranuclear U1-70K-ir neurons in the FC layer V across the two groups. p<0.05.*

spliceosome with highly likelihoods of widespread mRNA splicing errors/disruptions in both cortices in DS. In addition we found a significant reduction of SRSF2 nuclear size in FC pyramidal neurons in layer V in DSD+ (Figure 3), an increase in FC 90 kDa SRSF2 proteins levels in DSD+ that strongly correlated with 4Rtau protein levels in the FC across the two DS groups, establishing an association between SRSF2 and alternative splicing of exon 10 of tau.

**Aim 2:** Likewise, SR proteins are controlled by reversible phosphorylation by CDC-like (CLKs) kinases and phosphatases. Specifically, CDC2-like kinases CLK1, 2, 3 and 4 have been associated with the alternative splicing of the exon 10 (Hartmann et al., 2001), suggesting that alterations in CDC2-like kinases via SR phosphorylation, perhaps acting on SRSF2 may affect NFT pathology. Nanostring data showed a dysregulation in the expression of several splicing and transcription-related genes in the FC between DSD+ and DSD- [CLK1, a serine-arginine kinase CLK1 involved in alternative splicing; MNAT1 gene: an assembly factor of the CDK-activating kinase, which is involved in transcription activation; TAF4 and TAF9, TATA-box binding protein associated factor 4 and 9, respectively, both involved in transcription activation by RNA polymerase II] (Figure 4a). Our attention was drawn to CLK1 gene, since we found a significant upregulation of CLK1 transcripts in the FC and in Pr in DSD+ compared to DSD- (Figure 4b). Western blot data confirmed the increase of CLK1 levels in the FC in DSD+ (Figure 5).

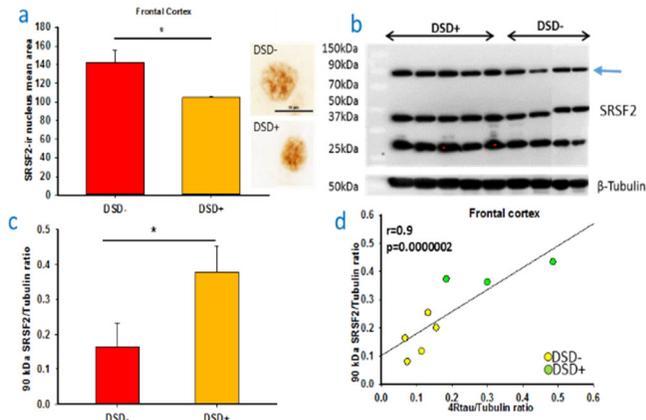


Figure 3. SRSF2 is association with 4Rtau in DS. a, Shows a significant reduction in nuclear size for SRSF2 in DSD+ compared to DSD-. b and c, a significant increase in FC 90 kDa SRSF2 proteins (blue arrow) levels in DSD+ compared to DSD- and c, a strong correlation between 90 kDa SRSF2 and 4Rtau protein levels in DS \* $p < 0.05$ . Scale bar = 10 $\mu$ m.

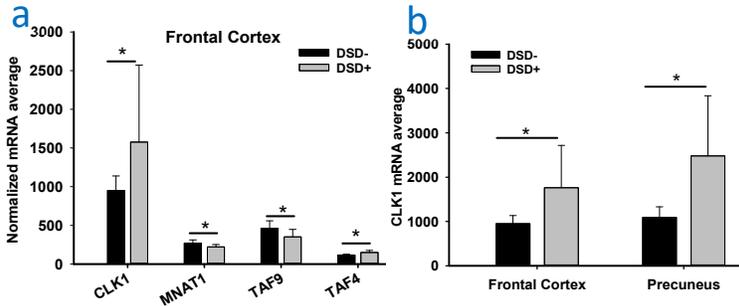


Figure 4. Preliminary FC nanostring data shows differential gene expression for several splicing and transcription factors between DSD+ and DSD-, including CLK1 (a). b, CLK1 mRNA expression was significantly increased in FC as well as in Pr in DSD+ compared to DSD-. \* $p < 0.05$ .

the CDK-activating kinase, which is involved in transcription activation; TAF4 and TAF9, TATA-box binding protein associated factor 4 and 9, respectively, both involved in transcription activation by RNA polymerase II] (Figure 4a). Our attention was drawn to CLK1 gene, since we found a significant upregulation of CLK1 transcripts in the FC and in Pr in DSD+ compared to DSD- (Figure 4b). Western blot data confirmed the increase of CLK1 levels in the FC in DSD+ (Figure 5).

**Aim 3:** Nuclear TBR1 immunofluorescence was observed in MAP2 positive neurons in disomic and trisomic hiPSC-CN cultures, indicative of hiPSCs differentiation into cortical V and VI lamina neurons. These TBR1 positive neurons were also positive for the nuclear splicing U1-70k marker. The presence of the nuclear splicing marker U1-70k in disomic and trisomic hiPSC-CN was independent of treatment, while the nuclear hnRNP A2B1 immunofluorescence was immunodetected in MAP2 positive neurons only in disomic, but not in trisomic, in both A $\beta_{25-35}$  treated and non-treated hiPSCs-CN cultures. While, mostly of the MAP2 positive neurons showed

nuclear SRSF2 immunofluorescence in disomic and trisomic A $\beta_{25-35}$  hiPSCs-CN cultures, nuclear SRRM2 immunofluorescence was observed in MAP2 positive neurons containing high expression of the phosphorylated AT8 tau marker in both A $\beta$  treated and non-treated cultures. In general, all phosphorylated AT8 and R3 tau positive cells were MAP2 positive in A $\beta_{25-35}$  treated and non-treated disomic and trisomic hiPSCs-CN cultures and expression was more pronounced in the cellular processes and soma in both disomic and trisomic hiPSCs-CN cultures treated with A $\beta_{25-35}$ . **These data was presented as a poster at Society for Neuroscience last year in Chicago.** Altogether, this data provide evidences that splicing, in particular alternative splicing, is highly compromised in DSD+ and key to advanced tau pathology described in the FC (Perez et al., 2019). These findings are foundation of our R01 that will be submitting on June 5<sup>th</sup>. As result of this project we have established a collaboration with Dr. Van Keuren-Jensen, from Tgen. And together will performed a broader analysis of transcript dysregulation and its relationship with tau pathology in the FC and Pr in DS using RNA sequence.

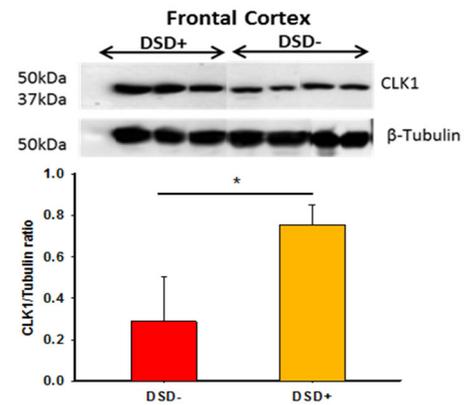


Figure 5. CLK1 proteins levels were significantly increased in FC of DSD+ compared to DSD-. \* $p < 0.05$ .

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Targeting to alpha7-nAChRs for controlling epilepsy in Alzheimer's disease.** Jie Wu, MD, PhD, Barrow Neurological Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

In Alzheimer's disease (AD) patients, the incidence of epilepsy is significantly higher than that in age-matched, non-AD controls<sup>1-2</sup>. Epilepsy is an important phenotype of AD<sup>1</sup>, and A $\beta$  accumulation induces aberrant neuronal hyperexcitation could be a primary upstream mechanism leading to cognitive deficits both in humans and animal models<sup>2,3</sup>. However, the mechanisms of the neural hyper-excitation in AD are largely unknown. Recently, we demonstrate that  $\alpha$ 7 nicotinic acetylcholine receptors ( $\alpha$ 7-nAChRs) play an important role in mediated chronic A $\beta$  exposure-induced neural hyper-excitation in primary hippocampal cultured neurons<sup>4,5</sup>. In this pilot proposal, we will extend our study to evaluate the roles of  $\alpha$ 7-nAChRs in hippocampal neural-network hyper-synchronization using hippocampal slices. We will test a novel central hypothesis that  **$\alpha$ 7-nAChR is an important target for neural hyper-synchronization and neuro-pathogenesis in AD models**. We propose a specific aim to test this central hypothesis.

### **Background and Significance:**

Alzheimer's disease (AD) is a dementing, neurodegenerative disorder characterized by increased accumulation of A $\beta$ , degeneration of neurons of basal forebrain, hippocampus and neocortex, and leads to a gradually-developing learning and memory deficit. Emerging evidence suggests that A $\beta$ -induced neural hyper-excitation contributes to neuro-pathogenesis of AD, but the mechanisms remain unclear. Recently, we demonstrate that  $\alpha$ 7 nicotinic acetylcholine receptors ( $\alpha$ 7-nAChRs) play an important role in mediated chronic A $\beta$  exposure-induced neural hyper-excitation in primary hippocampal cultured neurons. The long-term goal of this project is to understand the neuronal processes associated with the early etiology of Alzheimer's disease (AD). Specifically, this project seeks to understand the functional modulation of a unique subtype of  $\alpha$ 7 subunit-containing nicotinic acetylcholine receptors (nAChRs) by amyloid beta (A $\beta$ <sub>1-42</sub>). AD is a neurodegenerative disorder characterized by the accumulation of A-beta<sub>1-42</sub>, early neuronal degeneration and development of learning and memory deficits. Recent findings have raised the possibility that neuronal hyperexcitability represents a primary upstream mechanism that may contribute to early cognitive deficits. Additionally, it has been postulated that chronically high levels of A-beta<sub>1-42</sub> may contribute to neuronal hyperexcitability (Dr. David Holtzman, personal communication) and subsequent basal forebrain and hippocampal neuronal degeneration leading to AD. In many brain regions homomeric  $\alpha$ 7-nAChRs mediate cholinergic signaling. Given the potent blockade of  $\alpha$ 7-nAChRs by A $\beta$ <sub>1-42</sub>, it is not clear if A-beta<sub>1-42</sub> induces neuronal hyperexcitability through compensatory upregulation of nAChRs and, in turn, neurodegeneration. This project seeks to test the hypotheses that chronic A-beta<sub>1-42</sub> leads to  $\alpha$ 7-nAChR up-regulation and subsequent neurotoxicity of basal forebrain and hippocampal neurons.

### **Preliminary Data, Experimental Design and Methods:**

**Experiment 1.** Comparing hippocampal  $\theta$  oscillations between WT and APP mice. We will compare hippocampal  $\theta$  oscillations between age-matched WT and APP mice using hippocampal slices.

**Experiment 2.** Determining the effects of  $\alpha$ 7-nAChR blockers on hippocampal  $\theta$  oscillations in WT and APP mice. In this experiment,  $\alpha$ 7-nAChR blockers (e.g., MLA 10 mg/kg, i.p.) will be injected to 10-month-old 3xTgAD mice for 7 days, then hippocampal  $\theta$  oscillations will be measured and

compare to AD mice without MLA treatment. WT mice will be used as a control experiment. In addition, we also will examine acute effects of MLA (1  $\mu$ M with 10-min pre-treatment) on hippocampal  $\theta$  oscillations in APP mice.

**Experiment 3.** Examining hippocampal  $\theta$  oscillations in WT mice treated with A $\beta$ . This experiment will follow the report from Dr. Selkoe's lab, in which the aggregated (oligomer) A $\beta$  will be microinjection into mouse hippocampus<sup>10</sup>. In this AD model, the hippocampal synaptic function and plasticity are impaired. We will examine hippocampal  $\theta$  oscillations in this AD model and compare these to the saline injected mice.

**Experiment 4.** Examining hippocampal  $\theta$  oscillations in  $\alpha 7^{-/-}$  mice treated with A $\beta$ . We will perform the same experiment as experiment 3, but use  $\alpha 7^{-/-}$  mice (10-month-old).

**Experiment 5.** Effects of  $\alpha 7$ -nAChR blockers on  $\theta$  oscillations in WT mice treated with A $\beta$ . This experiment will be designed to exclude other nAChR subunits compensations in  $\alpha 7^{-/-}$  mice.  $\alpha 7$ -nAChR blockers (e.g., MLA 10 mg/kg, i.p.) will be injected to 10-month-old  $\alpha 7^{-/-}$  mice for 7 days, then hippocampal  $\theta$  oscillations will be measured, which will be compared to  $\alpha 7^{-/-}$  mice treated with A $\beta$  but no treatment with MLA.

### **Proposed One-Year and Long-Term Outcomes:**

From these experiments, we expect to collect novel data to show (1) APP mice exhibit hyper-synchronization compared to WT mice, (2)  $\alpha 7$ -nAChR blockers (treated *in vivo* and *in vitro*) prevent APP mice hyper-synchronization, (3) Micro-injection of A $\beta$  into hippocampus in WT mice enables to induce neural hyper-synchronization, (4)  $\alpha 7$ -nAChR blockers (treated *in vivo* and *in vitro*) prevent A $\beta$ -induced hyper-synchronization, and (5) Micro-injection of A $\beta$  into hippocampus in  $\alpha 7^{-/-}$  mice fails to induce neural hyper-synchronization. All experimental approaches for this proposal are routinely performed in PI's lab. Hippocampal  $\theta$  oscillations, as a biomarker of neural-network synchronization, will be induced by chemical (50  $\mu$ M carbachol) and measured using field potential recordings (single channel or multi-channel recording systems).

Collectively, this is a highly **significant** pilot proposal, extended from our previous project, which demonstrates a novel mechanism that may underlie neuronal hyper-synchronization in AD. By understanding the **impact of  $\alpha 7$ -nAChRs in A $\beta$ -induced neural hyper-synchronization in AD models**, we would not only help to explain the targets and mechanisms of epileptogenesis in AD, but also leverage as a new strategy for treating AD.

### **Year End Progress Summary:**

Since 07/01/19 to current (03/01/20), experiments have been running successfully. We have finished designed experiments 1-3. We summary experimental progress as followings:

**Experiment 1.** Comparing hippocampal  $\theta$  oscillations between WT and APP mice. We will compare hippocampal  $\theta$  oscillations between age-matched WT and APP mice using hippocampal slices.

**Progress:** We have compared hippocampal  $\theta$  oscillations between age-matched WT and APP mice using hippocampal slices preparation. We used field potential recordings in hippocampal slices. Hippocampal theta oscillations were induced by cholinergic agonist (50  $\mu$ M carbachol for 30-40 min). We have collected 10 data of hippocampal slices from either 5-6 WT or APP mice. Our results showed that the hippocampal theta oscillations occurred more obviously in APP mice than that in WT mice. These results suggest that APP mice exhibit hyper-synchronization, which support our working hypothesis.

**Experiment 2.** Determining the effects of  $\alpha 7$ -nAChR blockers on hippocampal  $\theta$  oscillations in WT and APP mice. In this experiment,  $\alpha 7$ -nAChR blockers (e.g., MLA 10 mg/kg, i.p.) will be injected to 10-month-old 3xTgAD mice for 7 days, then hippocampal  $\theta$  oscillations will be measured and compare to AD mice without MLA treatment. WT mice will be used as a control experiment. In

addition, we also will examine acute effects of MLA (1  $\mu$ M with 10-min pre-treatment) on hippocampal  $\theta$  oscillations in APP mice.

**Progress:** We have performed several experiments. First, we compared hippocampal theta oscillation between WT and  $\alpha 7$ -nCAhR KO mice, and found that  $\alpha 7$ -nCAhR KO mice showed weaker hippocampal  $\theta$  oscillations, suggesting that hippocampal  $\alpha 7$ -nCAhRs play a role in modulation of the CCh-induced hippocampal  $\theta$  oscillations. Second, we have examined acute effects of  $\alpha 7$ -nAChR blocker (MLA 1  $\mu$ M with 10 min pre-treatment) on the CCh-induced hippocampal  $\theta$  oscillations in WT mice, and found that bath-application of MLA reduced the CCh-induced hippocampal  $\theta$  oscillations.

**Experiment 3.** Examining hippocampal  $\theta$  oscillations in WT mice treated with A $\beta$ . This experiment will follow the report from Dr. Selkoe's lab, in which the aggregated (oligomer) A $\beta$  will be microinjection into mouse hippocampus<sup>10</sup>. In this AD model, the hippocampal synaptic function and plasticity are impaired. We will examine hippocampal  $\theta$  oscillations in this AD model and compare these to the saline injected mice.

**Progress:** We have completed these experiments. We have successfully established *in vivo* A $\beta$  toxicity model by microinjection of oligomeric A $\beta_{1-42}$  (100 nM) into WT hippocampus. We have performed two series of experiments using this *in vivo* A $\beta$  toxicity model. First, we examined animal learning and memory behavior (Water Maze), and found that after injection of oligomeric A $\beta_{1-42}$  into WT hippocampus for 1 week, mice showed a deficit of learning and memory behavior compare to the mice received injection of saline into hippocampus. Then, we examined hippocampal electrophysiology using this *in vivo* A $\beta$  toxicity model. In hippocampal slices prepared from treated (microinjection of oligomeric A $\beta_{1-42}$ ) or from control (microinjection of saline) mice, we measured hippocampal CA1 tetanic LTP (theta burst stimulation, 100 Hz 5 trans with 20 ms interval) and the CCh-induced hippocampal  $\theta$  oscillations, and found an impaired hippocampal LTP in treated group compared to control group. These results suggest that the microinjection of oligomeric A $\beta_{1-42}$  (100 nM) into WT hippocampus can establish a reliable *in vivo* A $\beta$  toxicity model, and we can use this model to evaluate the impact of  $\alpha 7$ -nAChR antagonist in neuronal protection against A $\beta$  toxicity in **Experiment 4 and 5**.

#### **Prediction of outcomes:**

Based on the data we collected from past 8 months, we predict that  $\alpha 7$ -nAChRs play an important role in mediated chronic A $\beta$  exposure-induced neural hyper-synchronizations, neurotoxicity and animal learning and memory deficits. We anticipate writing a research paper by the end of this year, and submit a new NIH R21 proposal by October 15, 2020.

**Project Progress Reports**  
**Critical Path Institute**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Training and Dissemination of Model-Informed Drug Development and Regulatory-Compliant Data Management in Alzheimer's Research for the AAC Community.** Jackson Burton, PhD, Nathan Hanan PhD, Yashmin Karten, PhD, Klaus Romero, MD, Sudhir Sivakumaran, PhD, Critical Path Institute; Arizona Alzheimer's Consortium

### **Specific Aims:**

- 1) Develop a hands-on training course for the use of regulatory-endorsed clinical trial simulators for Alzheimer disease and pre-dementia.
- 2) Develop a hands-on training course on the implementation of regulatory-grade data standards for non-clinical and clinical research in Alzheimer disease.

### **Background and Significance:**

Investment in AD research could be negatively impacted by recent high-profile drug development failures. Despite a continuously growing understanding of disease biology and drug mechanisms of action, further progress in model-informed strategies is needed to continue advancements in Alzheimer's drug development. Innovations in clinical trial design quantitative approaches could help bring effective treatment options to AD patients faster by accelerating development of effective new drugs and reducing failure rates in expensive late-phase development.

In AD, exploring more informative and predictive endpoints to assess treatment response has become an active area of research. Alternative metrics that require shorter periods of observation or provide more precise assessment of treatment effects could lead to more rapid completion of clinical trials and require fewer patients. Promising among these alternative metrics are model-based metrics, such as those based on longitudinal continuous ADAS-Cog and CDR-SB measurements. There is growing interest on the part of FDA in using model-informed approaches to help balance the risks and benefits of AD drug development by identifying optimal trial design approaches and broad stakeholder engagement and discussion around this topic can be beneficial [3-5].

The adoption of regulatory-grade data standards, such as the AD CDISC V2.0 data standard, serves as a platform to catalyze reproducible research, data integration, and efficiencies in clinical trials. It allows for the mapping and integration of patient-level data and provides a foundation for future studies, data sharing, and long-term registries in AD. The availability of consensus data standards for AD has the potential to facilitate the initiation of clinical studies and increase sharing and aggregation of data across observational studies and among clinical trials, thereby improving our understanding of disease progression and treatment [6].

However, there are regulatory-grade standards with application in AD research, which also carry importance in catalyze reproducible research, data integration, and efficiencies in clinical trials. They allow for the mapping and integration of experiment-level data.

Training on the use of these tools is critically important for AD researchers in the 21st Century. Of particular significance, these training opportunities will foster model-informed study design, as well as data interoperability, which can make AAC a leader in the adoption and application of these solutions.

## **Preliminary Data, Experimental Design and Methods:**

### **Preliminary Data**

A clinical trial simulation (CTS) tool was developed to describe disease progression based on longitudinal Alzheimer's Disease Assessment Scale-Cognitive sub-scale (ADAS-Cog) scores in mild-to-moderate AD, in a three-stage approach: (1) construction of a standardized database, (2) model development and evaluation, (3) FDA/EMA review for endorsement. To capture the maximum amount of information available for development of the CTS tool, data from a variety of sources were needed, requiring a model that simultaneously fitted summary and patient-level data. The CAMD database consists of patient-level, control-arm clinical trial data (both on stable background therapy and placebo only) from CPAD members. Demographics, genetics, and individual items from cognitive scales (MMSE, ADAS-Cog, etc.) were included.

Based on the CPAD database, an equivalent CTS tool was developed for pre-dementia, using CDR-SB as the endpoint. The model accounted for baseline intracranial volume corrected hippocampal volume (ICV-HV), APOE-ε4 carrier status, baseline MMSE scores, baseline CDR-SB, baseline age and sex as relevant covariates. The CTS tool allows the user to perform simulations to estimate sample size and statistical power; enrichment strategies can be evaluated under different assumptions and trial design options. Together with the range of MMSE scores at baseline and the proportion of APOE-ε4 carriers, the most appropriate ICV-HV threshold can be selected to increase the likelihood of demonstrating drug effects in aMCI clinical trials.

### **Experimental Designs and Methods**

The proposed training course on the use of clinical trial simulators for mild-to-moderate AD and pre-dementia will be a hands-on training opportunity, aimed at researchers of all backgrounds, regardless of their coding abilities in specific statistical packages like R, STAN or NONMEM. The training will be focused on the user-friendly graphical user interfaces.

The development of the didactic content for the course will be carried out by C-Path's Quantitative Medicine Team. The didactic content will start with an overview of the quantitative machinery behind the simulators, followed by an overview of the simulation environment of the GUI, and how to interpret the simulation outputs. This will be followed by case studies focused on specific and realistic study design challenges for drug development and observational studies, ranging from the definition of entry criteria, enrichment strategies, stratification approaches, sample size estimations, balancing of power and attrition, definition of hypotheses for statistical inference and their impact on analysis approaches, definition of frequency of observations, as well as total-follow-up time. These specific case studies will be developed to highlight relevant challenges to be addressed through the use of the regulatory-endorsed clinical trial simulators, aiming for the attendees to execute trial simulations, followed by a discussion of individual results and interpretation, based on the simulation outputs.

The proposed training course on the adoption and implementation of regulatory-grade data standards for non-clinical and clinical research will be a hands-on training opportunity, aimed at researchers of all backgrounds, regardless of their knowledge of CDISC, SDTM, SEND or ADAM standards. The training will be focused on the annotation of CRFs and laboratory notebooks, as well as on basic concepts of relational table data structures, and the generation of analysis subsets.

The development of the didactic content for the course will be carried out by C-Path's Data Collaboration Center (DCC). The didactic content will start with an overview of the logic behind standard terminology, followed by an overview of standard data structures for non-clinical and clinical data. This will be followed by case studies focused on specific and realistic scenarios for case report form annotation challenges, followed by case studies focused on specific and realistic

scenarios for relational table structures, and their relationship with the generation of analysis subsets.

**Proposed One-Year and Long-Term Outcomes:**

One-year outcomes: Train at least ten researchers. Present the results of this training effort at the American Conference on Pharmacometrics.

Long-term outcomes: Make AAC a leader in the adoption and application of quantitative-based study design, and data interoperability.

**Year End Progress Summary:**

The training covered the components of the CTS tools, including ADAS-Cog and CDR-SB as the endpoints, as well as the covariates that may affect disease progression, such as baseline hippocampal volume, baseline age, sex, APOE4 status, and baseline severity (baseline MMSE). The training also covered findings from the modeling effort, such as the quantitative estimates for APOE4 carriers showing a faster rate of progression than noncarriers, as well as providing a quantitative estimate supporting current thinking about risk factors, with younger patients progressing faster. The training also covered the model components providing a platform that enables simulation of a wide range of clinical trials according to variations in (1) drug, (2) disease state, and (3) trial design, to select a trial design with a high likelihood of detecting a treatment effect as well as to evaluate the trade-off between sample size and power. The technical and scientific intricacies of the CTS tool are described in more detail (see 2018-2019 CPAD AAC progress report) The CTS tool's flexibility to simulate beyond the standard parallel design used in most phase II and III AD clinical trials is important given that drugs may exhibit different nonlinear effects was also covered. Finally, the training covered how clinical trial simulations based on the models demonstrate that the inclusion of aMCI subjects with baseline ICV-HV below the 84th or 50th percentile allowed an approximate reduction in trial size of at least 26% and 55%, respectively. The training was successfully attended by 23 industry researchers, 14 CRO scientists, three government researchers and two academic scholars. Feedback received from attendees was overwhelmingly positive, with a request to consider including more technical background in non-linear mixed effects modeling, which will be incorporated into future training offerings.

**Project Progress Reports**  
**Mayo Clinic Arizona**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Normal and Pathological Aging (Preclinical Alzheimer's Disease).** Richard J. Caselli, MD, Dona E.C. Locke, PhD. Mayo Clinic Arizona; Arizona Alzheimer's Consortium.

### **Project Description:**

Cognitively normal individuals age 21-99 (most age 45-70) undergo 1) APOE genotyping to categorize their relative risk for developing Alzheimer's disease; 2) longitudinal neuropsychological and behavioral assessments; and 3) serve to create a biorepository for DNA, serum, plasma, viable frozen lymphocytes, and immortalized cell lines to determine what factors divert individuals from normal to pathological aging/Alzheimer's disease with the intent of identifying optimal timing of treatment and new potential therapeutic targets for preventing this divergence (prevention of Alzheimer's disease). This "APOE Cohort" also serves as a core resource for multiple collaborative projects within our site and for the consortium.

### **Specific Aims:**

- A. To maintain and grow a unique cohort of human aging in which we characterize the effect of APOE gene dose (a risk factor for Alzheimer's disease) on age-related changes in:
  1. Mentation (neuropsychological measures of cognition and behavior; subjective assessments by observers and self; sleep parameters)
  2. Brain Imaging (structural brain changes [MRI], functional [FDG-PET], amyloid-PET, tau-PET)
- B. To correlate longitudinal changes on each of these measures with clinical outcomes (mild cognitive impairment, Alzheimer's dementia, non-Alzheimer's dementia)
- C. To characterize the influence of other demographic, genetic, epigenetic, and health factors on cognitive aging trajectories
- D. To create a biobank of serum, plasma, DNA, frozen viable lymphocytes, and immortalized cell lines of this cohort.
- E. To function as a core resource collaboratively supporting other investigators
- F. To support, where appropriate, activities of the NIA funded Arizona Alzheimer's Disease Center

### **Background and Significance:**

Even at the earliest clinical stages of Alzheimer's disease (AD), amyloid pathology has nearly peaked yet neither symptoms nor brain atrophy correlate well with amyloid burden. Failed anti-amyloid therapies have been blamed on being started too late, resulting in new disease modifying strategies that begin during the preclinical, asymptomatic stage. Our work to date has helped to define and characterize the preclinical stage of AD, differentiating normal from pathological aging. Themes of our current research include 1) identification of preclinical disease modifying attributes (genetic, medical, demographic, and others), 2) extension of preclinical testing and precision medicine into the clinical practice domain, and 3) integration of multiple data sources into predictive algorithms.

### **Preliminary Data:**

To date we have completed APOE genetic testing on over 2800 participants from which were selected our study population for further testing. We have completed one or more epochs of neuropsychological testing on 838 individuals (table) including 474 APOE e4 noncarriers, 256 e4 heterozygotes, and 103 e4 homozygotes (we await final APOE results on the remainder). We

have over 10,000 plasma and serum samples from 618 (over serial epochs in many cases), whole blood from 707 and DNA from 773 individuals. 497 have immortalized cell lines established including all of those who have had brain imaging. Among our many accomplishments, we established cognitive aging trajectories for each of 3 APOE genotypes (1-3), the differential impact of modifying factors such as cardiovascular risk factors (4) as well as personality factors (such as proneness to stress) (5,6) and subsequently have shown that pre-MCI deviates from normal aging roughly 20 years before incident MCI diagnosis (7).

<u>Epoch</u>	<u>n</u>	<u>months followup</u>
1	838	0
2	657	30.0 (13.2)
3	502	60.2 (20.3)
4	416	88.6 (26.4)
5	361	114.74 (30.3)
6	292	137.8 (29.9)
7	224	158.9 (32.5)
8	165	179.6 (28.5)
9	125	196.2 (27.2)
10	77	213.9 (24.2)
11	54	224.3 (23.4)
12	40	242.4 (22.3)
13	23	253.9 (20.8)
14	4	270 (21.5)

**Proposed One-Year and Long-Term Outcomes:** In addition to maintaining the ongoing evaluation of this important cohort, our goals for the next one year include:

1. Continue our genetic study of unexpectedly young onset dementia patients with whole exome sequencing and bioinformatics analysis of a large gene set encompassing identified risk genes for Alzheimer, disease, frontotemporal lobar degeneration, and Parkinson’s disease
2. Compare the longitudinal trajectories of FDG-PET and MRI volumetrics, with neuropsychological tests in patients with and without eventual progression to MCI and dementia to determine how long in advance of diagnosis imaging trajectories significantly deviate from the nonprogressor group and that compares with our newly established cognitive trajectories.
3. Continue to support our collaborative projects
4. Continue to maintain the shared plasma/serum biobank resource
5. Continue to strategically merge, where appropriate, data from this project with the NIA P30 Alzheimer’s Disease Center data to create a much larger dataset encompassing the entire adult lifespan and all stages of cognition including young adulthood, middle age, and elderly normal stratified by APOE genotype and other demographic properties, as well as MCI and dementia.

**Year End Progress Summary:**

1. We published the results of our cognitive and behavioral aging trajectories contrasting individuals who developed incident MCI with those remaining clinically normal and showed that the earliest cognitive changes predate incident MCI diagnosis by 20 years, rivalling the earliest biomarker changes and implying that current pathophysiological models which posit a linear sequence of change with cognition lagging are in need of revision (7).
2. Using a validated Autism Questionnaire, we showed for the first time that roughly 10% of non-selected participants in a cognitive aging study meet or exceed the criterion score that defines the broad autism phenotype, and that this contributes to escalating subjective cognitive impairment over age 65 in the absence of objective cognitive decline (relative to controls) (8).
3. For the first time we were able to directly assess personality change during the transition from presymptomatic to MCI and showed that it is characterized by increasing proneness to stress and decreasing openness to new ideas and actions preceding but providing a fertile substrate for the emergence of behavioral disorders (6).
4. We completed a major transition in biobanking with the transfer our DNA bank from Mayo Clinic Florida (Rademakers lab) to Mayo Clinic Arizona, and have made major revisions to biosample tracking to facilitate sharing of biospecimens.
5. In addition to supporting our existing collaborations, we established a new collaboration supporting an R01 grant submission with Dr. Rui Chang at the University of Arizona.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Predicting Cognitive Decline in Cognitively Unimpaired Individuals.** Cynthia M. Stonnington, MD. Mayo Clinic Arizona; Arizona Alzheimer's Consortium.

### **Project Description:**

Cognitively unimpaired individuals age 21-99 (most age 45-70) undergo 1) APOE genotyping to categorize their relative risk for developing Alzheimer's disease; 2) longitudinal neuropsychological and behavioral assessments; and 3) serve to create a biorepository for DNA, serum, plasma, viable frozen lymphocytes, and immortalized cell lines to determine what factors divert individuals from normal to pathological aging/Alzheimer's disease with the intent of identifying optimal timing of treatment and new potential therapeutic targets for preventing this divergence (prevention of Alzheimer's disease). This project will capitalize on the existing longitudinal data base of imaging, neuropsychological testing, and genetic testing to establish how a clinician might use a combination of such data to identify pre-clinical predictors of disease and to determine the probability of developing disease for any given individual patient.

### **Specific Aims:**

1. To identify participants in our longitudinal study of aging who have baseline imaging and have shown evidence of cognitive decline by having developed incident MCI.
2. To preprocess MRI scans using cortical thickness, i.e., Freesurfer, and grey matter volume, i.e., SPM, methods, as well as surface multivariate tensor-based morphometry (mTBM) and grey matter morphology signatures to study important structural MRI AD biomarkers. Compare region of interest and whole brain differences between decliners and nondecliners for each of the methods.
3. To develop methods to predict decline using FDG PET, MRI, amyloid imaging, genetic, and neuropsychological data by creating training sets of baseline data from participants with decline and from participants who have at least two epochs of data and show no decline.
  - a. Examine the statistical power in distinguishing the two groups using Receiver Operating Curve (ROC).
  - b. Examine prediction accuracy by using machine learning methods.
4. To evaluate additional genetic markers contributing to risk for cognitive decline, including BDNF Val66Met polymorphism.

### **Background and Significance:**

Even at the earliest clinical stages of Alzheimer's disease (AD), amyloid pathology has nearly peaked yet neither symptoms nor brain atrophy correlate well with amyloid burden. Anti-amyloid therapies have all fallen well short of expectations to date, for the generally held reason that they are started too late, and that for a disease modifying agent to be effective it must be started during an earlier, preclinical stage, i.e., before patients develop symptomatic memory loss. Preclinical AD is superficially indistinguishable from normal aging. We therefore plan to develop methods to differentiate normal from pathological aging by combining imaging-based biomarkers, neuropsychological, and genetic data to better identify those individuals on the cusp of symptoms and therefore most likely to benefit from treatment.

### **Preliminary Data.**

1. From a total of 139 ADNI participants who were diagnosed as MCI and had baseline FDG PET and MRI imaging data, 78 ( $75.8 \pm 7.0$  years old) developed incident AD during the subsequent 36 months, and the remaining ( $75.3 \pm 8.0$ ) did not during the same period. FDG PET measured glucose uptake, MRI measured hippocampal volume and ADAS-mod at baseline all distinguished MCI converters from non-converters, but, using ROC, the sensitivity and specificity showed increased statistical power when these modalities were combined (sensitivity=82%, and specificity=80%).
2. From our longitudinal APOE data base of cognitively normal individuals, we have identified 21 individuals with baseline FDG PET and MRI and neuropsychological data who subsequently developed incident MCI, along with 180 in the same age cohort who remain cognitively normal also had FDG PET and MRI and neuropsychological data.
3. From our longitudinal APOE data base of 180 cognitively normal individuals with baseline FDG PET and MRI and neuropsychological data, we have identified 18 who show evidence of cognitive decline but have not yet developed MCI or AD.
4. From our longitudinal APOE data base, we identified 14 individuals with amyloid imaging data who also had evidence of cognitive decline but remained cognitively normal and matched by age, sex, APOE status, and education to 14 individuals who did not show any cognitive decline. At  $P < .005$  (uncorrected), decliners had significantly greater evidence of fibrillar A $\beta$  burden in comparison to nondecliners (1)
5. From our prospective cohort study of aging, we examined baseline CMRgl, Pittsburgh B (PiB) PET measured amyloid burden, and subsequent rate of change in cognition from 114 CU adults (59 with PiB PET) who had been both BDNF and APOE4 genotyped. Among APOE4 carriers, BDNF Met carriers had significantly higher frontal CMRgl and slower decline of frontal CMRgl over time than the BDNF val/val group but no significant differences in decline of cognitive scores. The BDNF effects were not found among APOE4 noncarriers. Increased amyloid deposition was positively correlated with areas of greater cerebral metabolism.

### **Experimental Designs and Methods:**

From our ongoing, longitudinal normal and pathological aging study, identify: 1) all participants with baseline imaging exhibiting cognitive decline according to definitions used in our prior studies; and 2) all participants with baseline imaging who developed incident MCI.

Both the FDG PET and PiB PET Distribution Volume Ratio (DVR) baseline images will be coregistered to MRI baseline images, and the MRI Dartel normalization will be used to normalize the MRI and PET data. For PiB PET scan data, the well-known graphical analysis Logan method and an automatically labeled cerebellar region-of-interest will be used to compute parametric brain images of the PiB DVR, a measure of fibrillar A $\beta$  burden. Together with the effects of age and sex, partial volume effect corrected PET kernel matrices will be created separately for segmented grey matter, cortical thickness, Dartel normalized MRI and PET images, APOE e4 genotype, and cognitive test score data. Regions of interest will be determined from published data that used a data set independent of ours.

For mTBM methods we will segment each baseline MRI scan with FSL (<http://fsl.fmrib.ox.ac.uk/fsl/fslwiki>), parameterize the hippocampal and ventricle surfaces as described previously, and generate the surface multivariate morphometry statistics (MMS) consisting of mTBM and radial distance (RD).

Firstly, we will examine the statistical power in distinguishing the two groups using Receiver Operating Curve method. Secondly, we will apply machine learned decision trees to various sets of features from brain imaging, genetic, and neuropsychological data. We will then test diagnostic and prognostic performance using different maximum number of features. Specifically, we will construct a collection of overlapping patches on the surface as the initial

sparse coding dictionary. Stochastic Coordinate Coding will then be applied to learn a dictionary and sparse codes. We will use the max-pooling algorithm on the newly learned high-dimensional features to obtain a final set of low-dimensional features. Finally, an AdaBoost classifier will be applied to categorize aMCI and cognitively unimpaired individuals with 5-fold leave-one-out cross validation adopted to evaluate classification accuracy, sensitivity, specificity, positive and negative predictive values.

**Proposed One-Year and Long-Term Outcomes:**

Produce computerized systems capable of diagnosis or prognosis for individuals who are cognitively normal based on chains of reasoning that a clinician can evaluate.

**Year End Progress Summary:**

We previously described 79% sensitivity and 78% specificity prediction of imminent clinically significant decline using standard automated brain mapping algorithmic programs, binary logistic regression and leave-one-out procedures (*Stonnington CM, Chen Y, Savage CR, Lee W, Bauer Iij RJ, Sharieff S, Thiyyagura P, Alexander GE, Caselli RJ, Locke DEC, Reiman EM, Chen K (2018) Predicting Imminent Progression to Clinically Significant Memory Decline Using Volumetric MRI and FDG PET. J Alzheimers Dis 63:603-615*). Here, we aimed to improve prediction with the same data set but using hippocampal surface multivariate morphometry statistics (*Shi J, Thompson PM, Gutman B, Wang Y, Alzheimer's Disease Neuroimaging I (2013) Surface fluid registration of conformal representation: application to detect disease burden and genetic influence on hippocampus. Neuroimage 78:111-134*) combined with patch-based sparse coding algorithms. For the latter, we chose a random forest algorithm.

From our APOE cohort study in Arizona, 18 cognitively unimpaired adults who subsequently progressed to the clinically significant memory decline within 2 years (progressors) were matched for age, sex, education, and apolipoprotein E4 allele dose to 20 adults who remained cognitively unimpaired for at least 4 years after baseline visits (nonprogressors). The same inclusion criteria and methods were then applied to the Alzheimer's disease Neuroimaging Initiative (ADNI) data set, resulting in a sample of 18 progressors and 34 nonprogressors who were older and had a greater percentage of males and non e4 carriers than the Arizona participants. We achieved 95% prediction accuracy in the Arizona cohort and 90% accuracy in the ADNI cohort. Combining the two cohorts (36 progressors and 54 nonprogressors) achieved 85% prediction accuracy, 86% sensitivity, and 83% specificity (see tables below). We are completing this manuscript to submit for publication. Our findings suggest that sparse coding together with the surface multivariate morphometry may be applied to individual volumetric MRIs to predict imminent progression to clinically significant memory decline with great accuracy and target individuals who could benefit from preclinical interventions, such as Executive and Memory Support System Training for Cognitively Unimpaired Older Adults at Risk for Dementia.

Table 1. Experimental Results: Arizona Cohort

<i>Hippocampal</i>	<i>MMS</i>	<i>MMS_left</i>	<i>MMS_right</i>
<b>Accuracy</b>	<b>0.95</b>	0.76	0.66
<b>Sensitivity</b>	<b>0.94</b>	0.71	0.78
<b>Specificity</b>	<b>0.94</b>	0.83	0.39

Table 2. Experimental Results: ADNI Cohort

<i>Hippocampal</i>	<i>MMS</i>	<i>MMS_left</i>	<i>MMS_right</i>
<b>Accuracy</b>	<b>0.90</b>	0.72	0.65
<b>Sensitivity</b>	<b>0.87</b>	0.67	0.50
<b>Specificity</b>	<b>0.94</b>	0.33	0.06

Table 3. Experimental Results: Combined Cohorts

<i>Hippocampal</i>	<i>MMS</i>	<i>MMS_left</i>	<i>MMS_right</i>
<b>Accuracy</b>	<b>0.85</b>	0.75	0.80
<b>Sensitivity</b>	<b>0.86</b>	0.77	1.00
<b>Specificity</b>	<b>0.83</b>	0.70	0.50

Interaction between BDNF Val66Met and APOE4 on biomarkers of Alzheimer’s disease and cognitive decline: 114 cognitively unimpaired (CU) adults (mean age 56.85 years, 38% male) from our AZ APOE cohort study with longitudinal FDG PET imaging and cognitive measures were BDNF and APOE genotyped. Of these, 58 also had Pittsburgh B (PiB) PET imaging. We examined baseline CMRgl, PiB PET amyloid burden, CMRgl change over time, and rate of change in cognition over an average of 15 years.

Results: Among APOE4 carriers, BDNF Met carriers had significantly increased amyloid deposition and accelerated CMRgl decline in regions typically affected by AD, but without accompanying acceleration of cognitive decline and with higher baseline frontal CMRgl and slower frontal decline relative to the Val/Val group. The BDNF effects were not found among APOE4 non-carriers. As expected, APOE4 carriers had significantly higher frontal amyloid deposition, decreased CMRgl in a pattern similar to AD, and greater rate of cognitive decline than APOE4 non-carriers.

Conclusion: Our preliminary studies suggest that there is an interaction between BDNF Met and APOE4 on amyloid- $\beta$  plaque burden and longitudinal PET measurements of AD-related CMRgl decline in cognitively unimpaired late-middle-aged and older adults, and that any cognitive effects may be mitigated by compensatory increases in frontal brain activity—findings that would need to be confirmed in larger studies.

This study was recently submitted for publication.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Psychosocial Intervention Development for Those Living alone with Mild Cognitive Impairment.** David W. Coon, PhD, Dona Locke, PhD. Arizona State University; Mayo Clinic Arizona; Arizona Alzheimer's Consortium.

### **Specific Aims:**

To develop a psychosocial intervention to improve and maintain quality of life for patients who are living alone with Mild Cognitive Impairment.

### **Background and Significance:**

Approximately 15-20% of people age 65 and older have Mild Cognitive Impairment (MCI), a condition characterized by measurable changes in thinking abilities that are noticeable to both people with MCI and their family/friends. However, people with MCI can still carry out their everyday activities. A recent systematic review suggests that approximately 32% of people with MCI go on to develop Alzheimer's within 5 years (Ward, Tardiff, Dye, & Arrighi, 2013). Depression appears to be quite common among MCI patients (25% in community samples; 40% in clinical samples) (Ismail, Elbayoumi, & Fischer, 2017), and MCI patients have reported significantly lower psychological quality of life compared to their peers with normal cognitive functioning. Moreover, living alone with MCI appears to place these MCI patients at higher risk for poorer outcomes (Muangpaisan et al., 2008). To date, no evidence-based treatments have been identified that improve and maintain quality of life for people living alone with MCI. The two investigators for the proposed project run intervention programs for individuals diagnosed with MCI and/or early-stage dementia. Early-stage Partners in Care (EPIC), led by Dr. Coon as a partnership with ASU and the Alzheimer's Association, is a program focused on patients with early-stage dementia and their care partners. This group dyadic intervention includes education and skill-training workshops designed to reduced stress, enhance well-being, and help manage challenges by hearing the patient's voice in terms of care values and future care preferences. The HABIT Healthy Action to Benefit Independence and Thinking program, led by Dr. Locke at Mayo Clinic, is a cognitive rehab and brain wellness intervention for patients with MCI and a program partner. HABIT aims to support functioning, improved quality of life, and strengthen partnerships. EPIC and HABIT can be seen as companion programs as each involves different types of interventions. The HABIT program involves: (1) cognitive rehabilitation (2) support group for both patient and partner (3) wellness classes (4) cognitive exercise and (5) yoga. However, neither program is designed to support MCI patients who do not have someone to be their partner (e.g., individuals living alone with MCI with no local family members). Using our experiences with EPIC and HABIT as a frame, we want to respond to local and federal partner requests (e.g., the Alzheimer's Association, local Area Agencies on Aging, and the U.S. Administration for Community Living) to develop an intervention program for this population.

### **Preliminary Data:**

The proposed short-term outcomes for this phase of the project were to conduct additional focus groups and focused interviews with people living alone with MCI and the providers who assist this population, based on findings from previous work; conduct and review analyses on data from these focus groups and focused interviews; use these analyses to develop screening, interview, and intervention components for an intervention for people living alone with MCI; conduct and analyze the data from a small single arm pre-post feasibility and acceptability project implementing the intervention. In addition, the data analyses would yield both professional

presentations at meetings like the Gerontological Society of America, the American Society on Aging or American Psychological Association as well as the submission of the pilot results to venues like *The Gerontologist (Practice Concepts Section)*, *the Clinical Gerontologist*, or *Dementia*.

Subsequently, the PIs would submit either an R21 or an R01 in 2021 or 2022, depending on the pilot project's findings.

### **Year-End Progress Summary:**

Focus groups with health and social services providers working with older adults with memory concerns identified key issues to address in the development of the intervention protocol (recruitment, screening, assessment, and intervention development). Findings raised the critical need to capture participants “upstream” indicating that very few individuals have a diagnosis of MCI and that it is challenging to distinguish the “worried well” from those with MCI and those with early-stage dementia without comprehensive neuropsychological evaluation and information from a collateral informant. Some health care providers suggested revising the project's current recruitment and screening materials to capture seniors with “memory concerns” and add additional telephone screening steps (e.g., telephone versions of the MoCA in combination with the TICS based on the current scientific literature) to determine those with symptoms consistent with MCI. Logistical barriers for intervention participation were also raised ranging from transportation/driving concerns to scheduling conflicts for those still employed. All components of the HABIT and EPIC projects were viewed as valuable. Recruitment of MCI participants for the focus groups has proven more challenging and the emergence of COVID-19 has delayed any delivery of the intervention. The PIs are revising the IRB protocols to permit telephone and/or video conferencing focus groups for those with MCI to gather additional feedback on intervention alternatives including delivery through online platforms.

**Project Progress Reports**  
**Midwestern University**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Following the Evidence: The Role of Microbes in the Development of Alzheimer's Disease.**  
Garilyn Jentarra, PhD, T. Bucky Jones, PhD, Johana Vallejo, PhD, Doug Jones, PhD, Jason Kaufman, PhD, Weidang Li, MD, PhD, Fernando Gonzalez, PhD, Kathy Lawson, PhD, Vanthida Huang, PharmD, Pamela Potter, PhD, Tony Tullot, MD, Ashlesh Murthy, PhD. Midwestern University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

#### **Specific Aim 1:**

Determine LPS localization in brain tissue as well as levels of immunological molecules in brain, spleen, and liver. **We hypothesize that LPS will localize to areas of pathology. We further hypothesize there will be differences in the presence/levels of specific innate immunological molecules in brain, liver and spleen, correlating with microbial presence and pathological features such as amyloid plaques, neurofibrillary tangles and cerebral amyloid angiopathy.**

Aim 1.1: Stain subject brain tissue for LPS to identify the location of the LPS. Determine if the LPS staining is localized to specific structures/areas (perivascular, intracellular, etc.), and if it co-localizes with amyloid or tau pathology.

Aim 1.2: Determine levels of acute phase proteins and other innate immune molecules in matched brain, liver, and spleen of subjects. Correlate those levels with determined bacterial identifications and LPS/LTA concentrations.

#### **Specific Aim 2:**

Determine if microbial elements (DNA, LPS, and LTA) are present in brain tissue from 3xTg and APOE4 mice (plus control mice for each), and assess correlations with genotype and pathology. **We hypothesize that microbial products are present in the brain in the absence of overt infection and that their presence/levels will correlate with amyloid/tau pathology, and vary with genotype.**

Aim 2.1: Determine bacterial loads by 16S rRNA gene sequencing in brain tissue from 3xTg, APOE4, and control mice obtained under sterile conditions.

Aim 2.2: Determine levels of LPS, LTA, A $\beta$  and hyperphosphorylated tau protein (pTau) in brain tissue of 3xTg mice and control mice. Evaluate if LPS/LTA levels correlate with levels of A $\beta$  and pTau in 3xTg mice and controls. Determine levels of LPS and LTA in APOE4 mice and APOE3 control mice. Asses the effects of mouse genotype on presence of microbial elements.

### **Background and Significance:**

There have been repeated suggestions in the literature that infection with various microbes including bacteria, viruses, and fungi, could play a role in the development of AD pathology. Recent data also indicates that the amyloid-beta peptide (A $\beta$ <sub>(1-40 or 1-42)</sub>) is a very strong anti-microbial peptide. Given our preliminary data, and data published by others, there is good reason for the brain to produce an anti-microbial peptide as part of an innate defense system. Along with descriptions of microbes in AD brain tissue, the literature also contains a large body of work describing the chronic immunological responses typical of AD. Microglial activation and cytokine production have been widely described in AD. The complement system is activated in AD brain tissue and many acute phase reactants, commonly produced by the liver as part of an innate response to infection, are found in increased levels in AD brain tissue. This includes C-reactive protein (CRP) and fibrinogen/fibrin.

The proposed experiments build on our preliminary data and incorporate an assessment and correlation of host responses that may be directed toward the microbes and microbial products that we have detected. We will be extending some aspects of our human studies to brain tissue from mouse models of AD and AD risk. Results from these experiments could alter the way that AD is understood and lead to more effective treatments.

### **Preliminary Data, Experimental Design and Methods:**

We have evidence of the presence of hundreds of different bacteria (by 16S rRNA gene sequencing) in the brain superior frontal gyrus (SFG), inferior temporal gyrus (ITG), and basal pons (BP) of human subjects. ELISAs for LPS (gram negative bacterial walls) and LTA (gram positive bacterial walls) were run to test for the presence of these molecules in matched serum and PBS lysates of brain tissue (SFG, ITG, BP) from our subjects. While serum LPS results were significantly different between normal and AD subjects, this may be due to the frequently immune compromised agonal state of AD patients. LPS levels were very high and quite variable in brain lysates in all groups, and did not show significant differences by group. We noted a lack of correlation between serum and brain LPS levels that may imply that the LPS derives from resident bacteria in the brain. Brain LTA levels were also variable, but substantially lower than LPS levels, suggesting that bacteria in the brain may be primarily gram negative.

#### **Methods Aim 1:**

Subject Group Selection/Tissue: Tissue from age-matched male and female subjects was obtained from the BSHRI brain bank. Groups: Normal non-demented controls, high pathology controls (HPCs, who have AD pathology but no cognitive impairment), patients with mild cognitive impairment (MCI, who would likely to progress to AD over time), and AD patients ( $n=12/\text{group}$ ).

**Aim 1.1:** We will use sections from SFG tissue to assess localization of the LPS within the tissue. We will use antibodies to stain fixed brain sections for either LPS alone, or LPS in conjunction with amyloid plaques or neurofibrillary tangles.

**Aim 1.2:** Tissue lysates from each subject will be assessed for the presence/relative levels of molecules associated with host response to infection. We will target immune factors described in the literature as occurring at increased levels in AD brain tissue. We will look for correlations between the types of bacteria present, LPS/LTA levels, subject characteristics (age, sex, APOE status) and brain pathology. We will also evaluate immune molecules produced in the periphery.

#### **Methods Aim 2:**

3xTg, C57/BL6, APOE4, and APOE3 mice will be raised under normal environmental conditions and euthanized at 12-13 months of age. The brain will be dissected out and bisected for use in either 16S rRNA gene sequencing for detection of bacterial DNA or ELISA for detection of LPS, LTA, A $\beta$ , and pTau.

**Aim 2.1:** DNA extraction and purification: Brain tissue will be dissected from transgenic and control mice under sterile conditions. DNA extraction will be then be performed using the Qiagen Powersoil DNA Isolation Kit. The NEBNext Microbiome DNA Enrichment kit will remove most of the methylated mouse genomic DNA in each sample, to allow for more accurate analysis.

**Aim 2.2:** ELISAs: Mouse brain tissue homogenates will be made in PBS. LPS and LTA ELISAs as well as A $\beta_{40}$ , A $\beta_{42}$ , and pTau ELISAs will be run on the homogenates.

Data analysis: Dr. Amy Buros-Stein, the MWU biostatistician, will be advising on and performing appropriate statistical significance (by t-test or ANOVA where appropriate) as well as running correlation analysis for our data.

### **Proposed One-Year and Long-Term Outcomes:**

By the end of this funding period, we expect to have published the data from the 16S rRNA experiments on human tissues as well the data from LPS/LTA ELISAs and immunological analyses. Sequencing and ELISA data from the mouse models will be published by the end of 2020. We are planning an NIH R15 REAP proposal in June of 2019, which will leverage our

existing and upcoming data. We will also apply for funding to the national Alzheimer's Association in the spring of 2020.

### **Year End Progress Summary:**

#### **Aim 1 Progress:**

For Aim 1.1, SFG brain tissue slices (PFA fixed and paraffin embedded) from our human subjects were stained with an LPS antibody, which revealed a wide-spread, diffuse staining pattern. We subsequently attempted to validate the staining target of the antibody using positive control slides made from smears of PFA-fixed gram negative bacteria. Those slides did not stain well with the antibody and we have since acquired a new LPS antibody and are currently in the process of staining additional human tissues.

We have made significant progress in the completion of Aim 1.2. Specifically, we have measured ferritin and fibrinogen levels in brain lysates by Western blot, and noted differences between subject groups, as previously indicated in the literature. ELISAs for TNF-alpha from spleen lysates have been completed, as have ELISAs for CRP from liver lysates from the same subjects. We are in the process of assembling this data to look for correlations between levels of those molecules, as well as correlations to subject characteristics (age, APOE status, brain pathology, etc.). We will also be evaluating this data in the context of the LPS/LTA levels and the specific bacteria that were identified by 16S rRNA gene sequencing in these subjects.

As an add-on to this project, we formed a collaboration with Dr. Haiwei Gu from ASU. Dr. Gu performed metabolomics analysis on our tissue preparations and identified distinct abnormalities in metabolism of saturated fatty acids in AD subjects as well as subjects with MCI, revealing that metabolic changes occur very early in the disease process. We will be following up on this data, and have additional experiments and a June 2020 NIH grant proposal planned.

#### **Aim 2 Progress:**

The work proposed in Aim 2.1 is well underway. We have sterilely collected tissue from all of the planned APOE3 and APOE4 mice. Tissue collection from 3xTg and C57/BL6 will follow shortly. An opportunity arose to include germ-free mice in the analysis (via a collaboration with the Sangram Sisodia lab at the University of Chicago). Tissues from those mice have already been harvested. DNA extraction and enrichment for microbial sequences is in progress for all of the collected tissues. The germ-free mice will provide a critical negative control that has thus far been lacking in our experimental design.

We performed dissections of one hemisphere of the brain so that separate analysis from the cortex, hippocampus, olfactory bulbs, and cerebellum could be performed. We flash froze the other brain hemisphere so that follow up RNA work can be performed to determine if bacteria were active prior to sacrifice of the animals. We also collected fecal pellets and material from the cecum so that we could determine if bacteria in the brain may have originated from the gut. When DNA preparation is completed, samples will be sent for 16S rRNA gene sequencing.

Aim 2.2 progress: Mouse brain tissue is currently being collected from APOE3 and APOE4 mice, and collection from 3xTg and C57/BL6 mice is planned. As soon as the tissue collection is complete, PBS lysates will be created from all of the tissues so that ELISAs can be run. The ELISAs required for LPS analysis have already been acquired.

We expect that the work in Aim 2.1 will, in particular, provide solid information regarding whether it is somewhat normal for a mammal to have bacteria present in their brain. Germ-free mice should show no bacterial sequences aside from contaminants that we know are associated with lab reagents. This will allow us to be more certain which bacterial sequences are contaminants and which truly represent resident bacteria in the brain.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Diabetic obesity results in cognitive impairment: Evaluation of the relationship between inflammation and senescence in the gut-brain axis, and the response to exercise and genistein treatment.** Layla Al-Nakkash, PhD, Thomas Broderick, PhD, Minsub Shim, PhD. Midwestern University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1. Determine ability of genistein and exercise to improve the inflammatory state in HFS mice.
2. Determine the effects of genistein and exercise on Alzheimer's-like pathology and synaptic markers in HFS-fed mice.
3. Determine the impact of genistein and exercise on HFS-induced senescence.

### **Background and Significance:**

Obesity resulting from ingestion of high energy foods, such as a high-fat diet (HFD), is known to result in loss of learning and memory function. Hippocampal neurogenesis has been shown to be impaired following consumption of HFD, which is important since this region of the brain plays a role in learning and memory, specifically of flexible memory (the ability to use previously learned information in a new situation). Metabolic syndrome, a major contributor towards cardiovascular disease, T2D and insulin resistance and inflammation, are all risk factors for Alzheimer's disease (AD) and dementia. It has been postulated that, with such dietary habits, cognitive infarctions are associated with increased amyloid beta deposits and increased formation of neurofibrillary tangles. Clinically, in overweight women cognitive dysfunction is an eventual outcome and given the epidemic of obesity in the US, this proposed study is timely. However, underlying mechanisms remain to be determined. Accumulating evidence supports the connection between organismal aging and cellular senescence; numbers of tissue senescent cells increases with age. In addition, it has been shown that removal of senescent cells suppresses or reverses aging and extends the health span of mice. Since high fat/high sugar feeding has been shown to induce senescence in mice it is likely that that senescence contributes to obesity-associated neurocognitive decline. Genistein is a naturally occurring isoflavonic phytoestrogen found in high concentrations in soy products and reaches micromolar concentrations in the serum. In our previous studies, the optimal concentration of genistein fed to mice was 600 mg, which yields serum genistein levels in the low micromolar range akin to levels achievable in humans eating a diet containing a glass of soymilk/day. Thus, the concentration of genistein used in our diets is feasible clinically, and causes no side effects in our murine studies. The role of genistein and exercise in reversing hippocampal dysfunction and senescence clearly deserves attention. We predicted that genistein administration would have beneficial effects on systemic inflammation and gastrointestinal-brain health in the current study. Moderate exercise is commonly recommended by physicians to assist in reversing obesity, and importantly, exercise has been shown to improve hippocampal-dependent learning and memory in older individuals and to ameliorate some of the memory dysfunction in HFD female C37BL/6J mice.

### **Preliminary Data, Experimental Design and Methods:**

We utilized male and female *C57BL/6J* mice (purchased from Jax Labs, aged 4-weeks), acclimated for 1-week, and then fed them a high-fat (HF) diet (containing: 60% fat, 20% protein and 20% carbohydrate purchased from Dyets, Inc.) with 42g/L liquid sugar (HS, sucrose and fructose combined) for 12 weeks. Mice were randomly divided into five groups/sex: HFHS, HFHS+genistein, HFHS+exercise, HFHS+genistein+exercise and lean controls (n=5/group).

Exercise, genistein and HFHS supplementation was throughout the duration of the study. The genistein supplement was added to the high-fat diet (Dyets, Inc., Bethlehem, PA) at a concentration of 600 mg. We have shown that this concentration of genistein incorporated in the diet is sufficient to produce significant beneficial modifications in intestinal function and bone health. Exercise duration was set at 30 min/day for 5 days/week, for the study duration of 12 weeks. Exercise intensity was 12 meters/min (i.e., the same as the American Heart Association guidelines for 30 minutes of moderate activity, for a total of 150 minutes/week). Comparison of sex-dependent effects and variances of mechanism(s) of action are fundamental to our long-term research objectives. Moreover, NIH guidelines require studies to utilize sex-dependent comparisons of animal models, thus proposing sex-dependent mechanisms, along with convincing preliminary data in future grant applications will be key. Mice were euthanized and tissues harvested and maintained at -80°C until use for these studies.

### **Proposed One-Year and Long-Term Outcomes:**

We hypothesized that administration of genistein or exercise would improve outcomes in the HFS-fed mice. We predicted that both genistein supplementation combined with regular exercise would have additive beneficial effects. We hypothesized that male and female mice would respond differentially to genistein and exercise, likely via varied mechanisms.

The results obtained from this 1-year proposal will be included in an NIH R15 proposal. We predict the data obtained will provide focused, pathway-driven mechanistic directions for our future extramural grant submissions. Moreover, we predict two manuscript submissions will result from these combined data from the three aims.

### **Year End Progress Summary:**

#### **Aim1. Determine ability of genistein and exercise to improve the inflammatory state in HFS mice.**

We utilized high-sensitive cytokine multiplex kits (Millipore) to evaluate the influence of genistein and/or exercise on serum cytokine levels. We found that in males: IL-12, IL-2, TNF- $\alpha$ , MCP-1 were all significantly elevated by high fat/high sugar feeding compared to lean controls, and genistein alone decreased TNF- $\alpha$ , MCP-1 levels back to leans, and exercise alone decreased MCP-1 levels back to leans. We found no changes in any of the cytokines tested in female groups. We encountered a lot of variability in the high fat/high sugar male cytokine levels and thus we plan to run another assay to increase sample size. We are in the process of assessing cytokine expression via western blot from jejunum tissue collected from these mice. We find that while NF- $\kappa$ B levels in brain tissue are comparable in both male lean control groups and males fed high fat/high sugar diet, interestingly, NF- $\kappa$ B levels are significantly decreased by all treatments.

#### **Aim 2. Determine the effects of genistein and exercise on Alzheimer's-like pathology and synaptic markers in HFS-fed mice.**

Currently, we have found using western blot of homogenized whole brains that: 1) amyloid beta (A $\beta$ ) levels are increased in brain tissue from the high fat/high sugar fed male mice, and that genistein, and genistein+exercise combined significantly decreases A $\beta$  levels, 2) phosphorylated tau levels are similarly increased in brain tissue from the high fat/high sugar fed male mice, and genistein+exercise combined significantly decreases phosphorylated tau levels. We are currently evaluating several other key proteins of interest that are markers of amyloid production and degradation. We predict to have those data in the next several months.

**Aim 3. Determine the impact of genistein and exercise on HFS-induced senescence.** We are at the beginning stages of addressing this aim. We have RNA isolated from jejunum tissues from lean and high fat/high sugar fed mice, to first assess if there are changes in IL-6 and IL-8, known

to be highly expressed in senescent cells. Dependent upon whether or not we find changes between the high fat/high sugar fed mice and controls, all other treatment groups will be examined. Moreover, in the next few months we plan to section tissues, and stain for  $\beta$ -galactosidase (a marker for senescence), and reactive oxygen (ROS, known to be elevated by senescence).

Future grant applications, publications and collaborations that arose from the research:

We aim to submit an NIH R15 grant within the next two NIH cycles to determine sex-dependent mechanisms of action of genistein and/or exercise on the gut-brain axis. Publications: one publication is in preparation aiming to address the influence of genistein and/or exercise on Alzheimer's-related markers in the brain, and a second publication is in preparation aiming to address the role of genistein and/or exercise on inflammation and senescence. To maximize tissue use from these murine studies, we have set up a collaboration with faculty at Michigan State University to ascertain the influence of genistein and/or exercise on bone.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Exercise-mediated mitigation of cellular senescence as a peripheral control mechanism for Alzheimer's disease risk.** Minsub Shim, PhD, Thomas Broderick, PhD, Layla Al-Nakkash, PhD, Midwestern University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

Currently, there are two basic types of rodent models for Alzheimer's disease (AD): 1) the transgenic (over-producing human Amyloid Precursor Protein and associated secretases, tau, and ApoE) mice and 2) the spontaneous senescence-accelerated mice including SAMP8. While transgenic models of AD provided valuable insight into the mechanisms of development and progression of AD, they over-express genes with mutations seen in early-onset AD, which account for less than 5% of all AD cases. Therefore, a spontaneous model where the process is not dominated by the production of human mutant proteins may be more suitable for the investigation of sporadic AD, in which aging is the critical risk factor. We propose to use SAMP8 mice to test exercise as an intervention that systemically affects the rate of aging, thereby mitigating the risk of AD.

SAMP8 is a sub-strain of senescence-accelerated mice (SAM) originally generated by inbreeding of AKR/J mice. The main characteristic of SAM is normal development and maturity of reproductive function, followed by an early manifestation of age-related phenotypes. SAMP8 have a shorter life-span (9-12 months) and display age-related deficits in learning and memory. They also exhibit other early-aging phenotypes, which makes them useful for the study of other age-related alterations of body function such as sarcopenia, osteoporosis, cardiovascular dysfunction, renal inflammation, and intestinal villi degeneration.

### **1. To define exercise as a modulator of cellular senescence in SAMP8 mice**

Specific Aim 1 will test our hypothesis that exercise will decrease tissue accumulation of senescent cells. The levels of senescence markers in the tissues of SAMP8 mice with or without exercise will be analyzed.

### **2. To explore the effect of exercise on age-related changes in the brain of SAMP8 mice**

In Specific Aim 2, we hypothesize that exercise alleviates age-related pathological changes in the brain of SAMP8 mice.

### **Background and Significance:**

The number of Americans aged 65 and older was 46.2 million in 2014, representing 14.5% of the U.S. population or one in every seven Americans. By 2060, there will be an estimated 98 million older persons, which is more than twice their number in 2014. Further, the 65-and-older age group's share of the total population will rise to nearly 24 percent, almost a quarter of the population. Since aging is a critical risk factor in a variety of human pathologies including AD, determining the causal cellular and molecular processes that lead to functional decline and frailty is crucial for achieving a goal of "healthy aging".

Consistent with this idea, the NIH/NIA recently issued an RFA titled "*Geroscience Approaches to Alzheimer's Disease*" (RFA-AG-20-013). This initiative aims to test whether interventions known

to systemically affect the rate of aging are effective as modulators of the incidence, progression, etiology, and treatment of AD.

While studies have established the links between human progeroid syndromes and specific gene mutations, the causes of aging or age-related diseases remain elusive. However, increasing evidence supports the connection between cellular senescence and organismal aging. The number of senescent cells increases with age in mammalian tissues, and such cells have been found at sites of age-related pathologies such as osteoarthritis and atherosclerosis. Additionally, it has been shown that the removal of senescent cells in mice suppresses or reverses aging. Moreover, caloric restriction, which slows down aging in almost all species, decreases the number of senescent cells in the tissues of mice. We also recently have shown that cellular senescence is increased in our transgenic mice which exhibit early-aging phenotypes.

Growing evidence suggests that environmental factors play an important role in aging. However, there is a fundamental gap in understanding how environmental stimuli promote or suppress functional decline during aging. For example, the beneficial effects of exercise are well-known, but how exactly does physical activity improve the function of different tissues and organs in the body?

#### **Preliminary Data, Experimental Design and Methods:**

Twenty-four 8~9-week-old male and female SAMP8 mice (2 groups; sedentary vs. exercise, 6 males and 6 females/group) will be purchased from ENVIGO (former Harlan Laboratories). After one-week acclimation, the mice will be subjected to exercise training using a treadmill designed for mice (Exer 3/6, Columbus Instruments, Columbus OH, USA) until the termination of the experiment at 6 months of age. The exercise groups will be run on a treadmill for 30 minutes, 12 meters/min/5 days/week for the study duration of 16 weeks. The intensity of exercise training corresponds to an estimated maximal oxygen-carrying capacity of ~75% in mice. Before the start of the exercise protocol, mice will be initially acclimated to daily 10-minute exercise sessions for one week. Dr. Broderick's laboratory has an established protocol for mouse exercise and, thus, we do not anticipate problems in performing mouse exercise. However, although unlikely, if we encounter problems, we will consider using voluntary running wheels.

At the termination of the experiment, 6-month-old mice will be sacrificed and various tissues will be harvested. From as early as 4 months onward, SAMP8 mice exhibit aforementioned brain pathologies that increase in number and extent with age. The tissues will be processed for subsequent analysis of senescence markers and AD pathology as described in Specific Aims. The data will be analyzed by an appropriate statistical method in collaboration with Dr. Amy Buros-Stein at Northwestern University. The comparison will be made between same-sex. Before and during the experiment, the mice will be examined for signs of lesions and distress, and healthy mice of similar weight will be included in the experiment. In addition, the consistency of food and water will be regularly checked.

#### **Proposed One-Year and Long-Term Outcomes:**

This funding will enable the cross-disciplinary collaboration between MWU scientists, aiming at securing extramural funding. We plan to submit an NIH proposal in response to the aforementioned RFA or other funding opportunities. Additionally, we anticipate one or two manuscripts submitted to peer-reviewed journals. The findings from this study will be presented at Northwestern University's Kenneth A. Suarez Research Day, as well as at the Arizona Alzheimer's Consortium Annual Scientific Conference. We also plan to present our data at a national scientific meeting.

We will establish a breeding colony of SAMP8 mice here at MWU. Currently, faculty members at MWU are involved in the research projects on various aspects of aging including AD, cancer, diabetes, macular degeneration, cardiovascular physiology, bone metabolism, and telomere biology (<https://www.midwestern.edu/about/mwuresearch/azfacultytable.html>). Our project will bring SAMP8 mice, which could be a valuable resource for the research community at MWU. It would also provide opportunities for collaborative research and extramural funding.

Students involved in the research described in Specific Aims will be able to obtain a better understanding of AD, as well as the biology of aging. They will learn and master various molecular/cellular biology techniques described in Specific Aims. The students will also perform histopathological analysis of mouse tissues. Additionally, the students will improve their written and oral skills in scientific communication by participating in manuscript writing and presenting their findings in scientific meetings.

#### **Year End Progress Summary:**

- The purchase of SAMP8 mice was delayed because of the license agreement between ENVIGO and MWU, and the project was started in November 2019. We originally proposed the use of a treadmill designed for mice. However, we found that SAMP8 mice were very reluctant to run on the treadmill. It appears that their sole purpose is to escape, by either jumping out of the treadmill or squeezing their way out through the brushes at the back of the treadmill. Since this is likely to cause them to get injured, the running paradigm was switched to a voluntary wheel system. After 16 weeks of voluntary wheel exercise, tissue harvest was completed in February 2020.
- We are currently isolating RNA from tissues from both groups (i.e., sedentary and exercise) of SAMP8 mice. The isolated RNA will be used for qRT-PCR analysis of cellular senescence. Additionally, protein lysates and tissue sections will be prepared for analysis of senescence and AD pathology.
- We have established a breeding colony of SAMP8 mice. These mice are available for collaborative research on various aspects of aging.
- We plan to present the findings from this study at MWU's Kenneth A. Suarez Research Day, as well as at the Arizona Alzheimer's Consortium Annual Scientific Conference in 2021. We also plan to present our data at a national scientific meeting.
- We plan to submit an NIH R15 to determine the mechanism by which suppression of cellular senescence in peripheral tissues decreases the incidence and progression of AD. Additionally, we anticipate one or two manuscripts submitted to peer-reviewed journals.
- We also have set up collaborations with the faculties at Michigan State University and Auburn University.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Studies on the effects of the telomere protection protein RAP1 and the epsilon isoform of Glial Fibrillary Acidic Protein on amyloid peptides produced by gamma-secretase.** Mark J Swanson, PhD, Nancy S Bae, PhD. Midwestern University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

**Specific Aim 1.** Determine how RAP1 and GFAP $\epsilon$ , including mutants of each, affect A $\beta$  production by  $\gamma$ -secretase in a reconstituted yeast system.

**Specific Aim 2.** Determine the effects RAP1 and GFAP $\epsilon$  on A $\beta$  production using human cells.

### **Background and Significance:**

The human RAP1 protein was originally identified as part of a complex that protects the ends of chromosomes. More recently, RAP1 was shown to have non-telomeric functions as well. Our data indicate that RAP1 is predominantly cytoplasmic in cells that are oxidatively stressed. To understand the unique role(s) of RAP1 in the cytoplasm, a yeast two-hybrid screen was performed using RAP1 as a bait and a human fetal cDNA brain library as prey. We identified a specific isoform of glial fibrillary acid protein (GFAP), GFAP $\epsilon$  as an interacting protein of RAP1.

GFAP is an intermediate filament protein expressed by astrocytes. Altered GFAP expression is associated with a variety of neurological diseases, and an increase in GFAP will lead to senescence in astrocytes, which plays a key role in the chronic inflammation associated with Alzheimer's disease (AD). GFAP $\epsilon$  is the only isoform of GFAP that is capable of interacting with the presenilin protein, PS1, mutations of which are associated with familial AD. AD is an age-related neurodegenerative disease characterized by a number of morphological abnormalities including neurofibrillary tangles and amyloid plaques. Mutations in the genes coding for amyloid precursor protein (APP) and the presenilins (PS1 and PS2) are frequently seen in familial forms of AD. APP, a transmembrane protein, is cleaved to form amyloid beta (A $\beta$ ) proteins. APP is cleaved by  $\gamma$ -secretase to produce proteins of variable lengths, namely A $\beta$ 40 and A $\beta$ 42. The catalytic subunit of  $\gamma$ -secretase is the PS1. Higher levels of A $\beta$ 42 promote self-aggregation resulting in the formation of senile plaques. The goal of our research is to determine the effects of the interactions of GFAP $\epsilon$  and RAP1 on the activity of PS1 as part of  $\gamma$ -secretase.

*The research in this application is significant because it will provide insight into the mechanisms for the production of amyloid peptides that may lead to plaque formation, a hallmark of AD pathology.*

### **Preliminary Data, Experimental Design and Methods:**

**RAP1, GFAP epsilon and PS1 interact in the yeast 2-hybrid system.** The yeast two-hybrid (Y2H) system is a simple yeast genetic method for detecting protein-protein interactions. Using this system, we identified GFAP $\epsilon$  as an interacting protein of RAP1. Our data show that GFAP $\epsilon$ , RAP1 and PS1 interact in a cooperative manner. GFAP $\epsilon$  is polymorphic, and we determined that different allelic isoforms interact with RAP1 differently. In addition, we found that the human RAP1 protein interacts with GFAP $\epsilon$ , but ape RAP1 does not.

**RAP1, GFAP epsilon and PS1 expression in human cell lines.** Our data show that the neuroblastoma cell line SH-SY5Y and the glioblastoma cell line U251 simultaneously express APP, PS1, GFAP $\epsilon$  and RAP1, and thus these lines can be used in our studies.

**Proposed One-Year and Long-Term Outcomes:**

***Specific Aim 1. Determine how RAP1 and GFAP epsilon, including mutants of each, affect amyloid beta (A $\beta$ ) production by  $\gamma$ -secretase in a reconstituted yeast system.***

We will reconstitute  $\gamma$ -secretase in yeast cells. We will use a specially designed yeast strain to determine the overall activity of  $\gamma$ -secretase. In addition, we will use a polyacrylamide gel electrophoresis system with 8M urea to separate A $\beta$  peptides and measure the A $\beta$ 40:A $\beta$ 42 ratio. Our goal is to determine how RAP1 and GFAP $\epsilon$ , as well as variants and mutants of these proteins, will influence the activity of  $\gamma$ -secretase and A $\beta$  peptide production using the yeast system.

***Specific Aim 2. Determine the effects RAP1 and GFAP epsilon on amyloid beta (A $\beta$ ) production using human cells.***

Our goals are to determine 1) whether GFAP $\epsilon$  and RAP1 play a role in A $\beta$  production due to their interactions with one another and PS1, and 2) whether the variants of GFAP $\epsilon$  and RAP1 alter A $\beta$  production. To achieve these goals, we will first delete the GFAP $\epsilon$  and RAP1 genes in human neuronal cell lines using CRISPR/Cas9. Deletions will be made in the SH-SY5Y (neuroblastoma) and U251 (glioblastoma) human cell lines. In order to test the specific amino acid changes that occur in GFAP $\epsilon$  and RAP1 proteins, we will modify the GFAP $\epsilon$  and RAP1 genes and clone these modified sequences into human vectors for expression. We will examine the total levels of A $\beta$  by immunoblotting. We will also determine the A $\beta$ 40:A $\beta$ 42 ratio in these cells using ELISA.

***Specific Deliverables/Future Plans.***

If granted, we propose to utilize funds from the award to establish complementary yeast and human cell systems for the detection of  $\gamma$ -secretase activity and measure A $\beta$  peptide production.

**Year End Progress Summary:**

One of our main goals is to establish a yeast-based  $\gamma$ -secretase system to measure  $\gamma$ -secretase activity in the presence of RAP1, GFAP $\epsilon$  and variants of each. In addition, we plan to use this system in the future to identify additional human proteins that may influence  $\gamma$ -secretase activity. We chose two yeast strains available in the lab that are commonly used for yeast 2-hybrid screens as our host strains. These strains were modified to accommodate all of the human genes that need to be expressed for the system. Each strain has three different reporter genes that we can use to monitor  $\gamma$ -secretase activity. Currently, we are finalizing the creation of a modified APP that we can use as a target for  $\gamma$ -secretase. This gene contains a yeast secretory signal at the amino terminus that will allow the protein construct to be translated into the endoplasmic reticulum for incorporation into the plasma membrane. This is fused to C99, which is the fragment of APP generated by  $\beta$ -secretase that is cleaved into A $\beta$  peptides by  $\gamma$ -secretase. At the carboxyl terminus, the yeast Gal4 transcriptional activator protein is fused. The fusion protein will be expressed as an integral membrane protein, sequestering the Gal4 activator at the plasma membrane. When the fusion protein is cleaved by  $\gamma$ -secretase, A $\beta$  peptides will be released outside of the cell, and the Gal4 protein will go into the nucleus and activate the reporter genes. This fusion gene is being assembled with yeast sequences for chromosomal integration and a marker gene for selecting those successful integration events.

The human  $\gamma$ -secretase complex consists of four subunits, PS1, Anterior Pharynx-1 (Aph-1), Nicastrin (Nic) and presenilin enhancer 2 (Pen2). Genes encoding each of these proteins were amplified by polymerase chain reaction (PCR), cloned into sequencing vectors, verified by sequence analysis and cloned into yeast expression vectors. Due to the number of genes we need to express in the yeast, we used yeast vectors with bidirectional, constitutively active promoters to clone the  $\gamma$ -secretase subunits. In order to express RAP1 and GFAP $\epsilon$  in this system, we generated an additional vector with bidirectional, constitutively active promoters. All of the plasmids are ready, and we only need to get the C99-Gal4 construct into the chromosome to begin generating data from this system.

In addition to the base system described above, we have generated numerous variants for determining their effects on  $\gamma$ -secretase. We have generated all of the GFAP $\epsilon$  alleles that encode proteins varying at amino acid 426 (threonine, alanine and valine). We have generated a RAP1 mutant that has the same sequence as that of great apes. All of the variants are currently being cloned into the system vector.

We want to measure overall  $\gamma$ -secretase activity, but we are also interested to determine if the specific cleavages are affected. Although we can purchase A $\beta$  peptides as controls for our gels, we have created *E. coli* expression plasmids with A $\beta$ 40 and A $\beta$ 42 to allow us to make these controls at a minimal cost.

Our other main goal has been to use human cells for determining the effects of RAP1 and GFAP $\epsilon$  on A $\beta$  production. We have isolated DNA from the U251 and SH-SY5Y cell lines and performed PCR to determine the GFAP $\epsilon$  alleles present in each. U251 cells are homozygous for the threonine allele, and SH-SY5Y cells are heterozygous having a threonine allele and a valine allele. Currently, constructs are being generated for the deletion of RAP1 and GFAP $\epsilon$  from the chromosomes. We are also generating plasmids for the transient and stable transfection of these cell lines with RAP1 and GFAP $\epsilon$  variants.

**Project Progress Reports**  
**Northern Arizona University**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Toward establishing causality of the gut microbiome via the gut-brain axis in Alzheimer's Disease.** Emily K. Cope, PhD, J. Gregory Caporaso, PhD. Northern Arizona University; Arizona Alzheimer's Consortium.

### **Project Description:**

The human body is host to trillions of microorganisms, collectively termed the human microbiome. Recent technological advances have vastly expanded our understanding of the human microbiome, and it is becoming clear that the human microbiome impacts diverse aspects of health, including neurological health. There is increasing evidence that commensal host-associated microbiota can impact the brain and even host behavior [reviewed in [7,8]]. Mechanistically, this may occur through stimulation of the vagus nerve, through microbially produced metabolites entering the circulatory system and possibly crossing the blood-brain barrier, or through microbially produced metabolites stimulating the immune system, among other mechanisms. Specific to AD, an altered gut microbiome may increase neuroinflammation, a process central to AD progression. In addition, bacteria in the GI tract produce a significant amount of amyloids (aggregated, insoluble proteins exhibiting  $\beta$ -pleated sheet structures), LPS, or other pro-inflammatory metabolites that can prime the immune system during ageing and increase risk for AD development [9,10].

### **Background, Significance and Preliminary Data:**

Recent work in humans suggests the composition and diversity of the gut microbiome differs in individuals with AD. AD patients were characterized by decreased gut microbiome richness (i.e., number of different types of microbes that are present) and decreased abundance of the bacterial genus *Bifidobacterium* relative to age-matched healthy controls [11]. Work in transgenic mice supports the observation that gut microbiota differ in mice exhibiting AD pathology relative to wild-type mice, and illustrates a correlation between the gut microbiome composition and cerebral amyloid- $\beta$  peptide levels or aggregation[12,13]. While these results are promising, additional studies are required to determine potential causal relationships between gut microbiota alterations and AD pathology.

These studies have provided a framework for NAU to perform AD-related research. We have now established a colony of 60 wild-type B6129SF2/J and 70 3xTg-AD mice. Fresh fecal pellets were collected weekly for microbiome analysis and mice were sacrificed at 8, 24, and 52 weeks for analysis of amyloid- $\beta$  deposition in the hippocampus and metatranscriptomics. We observed a striking difference in the microbiome composition of our wild-type and 3xTg-AD mice between 3-12 weeks, and these differences remain apparent for the duration of our experiment. We observed 18 bacterial genera from 4 bacterial phyla that differ in their abundance between the wild-type and 3xTg-AD mice using ANCOM [14]. In subsequent studies, we will have the opportunity to determine whether these differences in microbiome composition may be causative of AD.

Gut microbiome composition in our mice changes predictably with time, such that using Random Forest machine learning regressors we can accurately predict the week that each sample comes from given only its microbiome composition (r-squared: 0.49; p=0.000002). When we work backwards from this model, we can identify specific taxa that are changing with time, and therefore with progression of 3xTg-AD mice through disease. In some cases, we observe bacterial taxa that change in abundance over time more so in 3xTg-AD mice relative to our non-AD wild type controls (e.g., *Turicibacter*). Other bacterial taxa change more over time in our wild

type mice (e.g., Lachnospiraceae), and some display similar longitudinal abundance profiles, though differing succession patterns, in both the wild-type and 3xTg-AD mice (e.g., *Clostridium*). These differences across our mouse strains point toward certain taxa that may be protective against AD progression (e.g., Lachnospiraceae) or causal of AD progression (e.g., *Turicibacter*). We are continuing to sequence and analyze longitudinal fecal samples from 3xTg-AD and wild-type mice as they age. We have implemented a fecal transplantation experiment to assess whether the differing microbiome states illustrated in Figure 1 has the ability to induce AD-like pathology in wild-type mice or lead to earlier disease progression in 3xTg-AD mice. We also aim to analyze the gut microbiota of single transgenic mice to determine host genetic factors that significantly contribute to gut microbiome composition. If successful, these results will clearly implicate the gut microbiome in AD.

### **Experimental Designs and Methods:**

We propose to longitudinally investigate the relationship between the gut microbiome, the gut inflammatory response, and the brain inflammatory response in triple transgenic mice on a mixed genetic background, which present with hallmarks of AD pathology including amyloid deposition and neurofibrillary tangles [3xTg-AD (B6;129-Tg(APP<sup>Swe</sup>,tau<sup>P301L</sup>)1Lfa *Psen1tm1Mpm/Mmjax*)] and wild-type B6129SF2/J mice. We have also included single transgenic mice APP-SWE Tg/Wt [B6;SJL-Tg(APP<sup>SWE</sup>)2576Kha)], which exhibit plaque formation, wild-type transgenic APP-Wt/Wt (B6;SJL-Tg(APP<sup>WT</sup>)2576Kha), and JPLN3(P301L) mice which exhibit tauopathy. Fecal samples will be collected weekly from mice for a total of 52 weeks. Mice will be sacrificed at 8, 24, and 52 weeks and the frontal cortex and hippocampus will be dissected and prepared for neuroinflammatory response and immunohistochemical analysis of amyloid- $\beta$  plaques tau protein as previously described [12]. Fecal microbiota transplants were performed from aged 3xTg-AD mice (12months) into young wild type and 3xTg-AD mice immediately post weaning. Briefly, fecal material was introduced via oral gavage daily for 5 days, then biweekly for 6 months.

### **Specific Aims:**

The primary goal of the proposed study is to evaluate changes in the fecal microbiota of transgenic mice and to determine, in single transgenic and 3xTg-AD mice, whether neuroinflammation, amyloid pathology, or tau pathology is increased when mice are given a fecal microbiome transplant (FMT) from aged 3xTg-AD mice. *The central hypothesis governing this proposal is that alterations in the gut microbiome contribute to neuroinflammation and AD pathologies in susceptible individuals.* Therefore, we propose the following Specific Aims:

*Specific Aim 1. To assess the longitudinal fecal microbiome in triple and single transgenic murine models representing separate tau and amyloid pathologies*

*Specific Aim 2. To evaluate the effect of fecal microbiota transplant (FMT) on progression of amyloid and tau pathology in 3xTg-AD and single transgenic mice.*

### **Proposed One-Year and Long-Term Outcomes:**

This project may ultimately lead to early markers (detectable through fecal samples) of amyloid- $\beta$  deposition or neurofibrillary tangles in pre-Alzheimer's patients, and to potential approaches to Alzheimer's prevention through alteration of the gut microbiome. In future studies, we plan to collaborate with AAC clinicians to obtain human fecal samples from patients with mild cognitive impairment and AD. We plan to submit an R01 to the NIA in October 2020 using results obtained in Years 1 and 2 of this study. We have identified an FOA that is well suited for the proposed research: PAR-19-070 (*Research on current topics in AD and related dementias*), which recently had a Notice of Special Interest (NOT-AG-19-012) published to investigate the role of pathogenic microbes in AD. We have recently submitted an R21 titled "Toward a mechanistic understanding

of the gut microbiota in Alzheimer’s Disease using quantitative Stable Isotope Probing” to the FOA PAR 19-071, which as a special interest NOT-AG-19-007.

**Year End Progress Summary:**

We have established a colony of wild-type B6129SF2/J, 3xTg-AD, APP-SWE tg/wt, APP-SWE wt/wt, and JPNL3 mice. Fresh fecal pellets are collected weekly for microbiome analysis and mice are sacrificed at 8, 24, and 52 weeks for analysis of amyloid-β deposition in the frontal cortex and metatranscriptomics (ongoing). We observed a continued difference in the composition of triple transgenic (3xTg-AD) mice over the course of one year. We observe 15 bacterial genera from 5 bacterial phyla that differ in their abundance between the wild-type and 3xTg-AD mice using ANCOM [14].

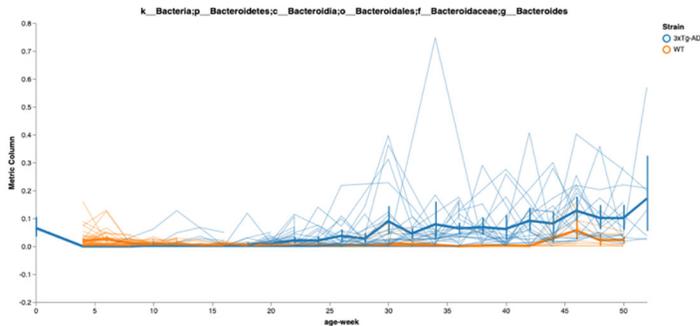


Figure 1. Random Forest regression demonstrating taxa that are predictive of age in mice. *Bacteroides* abundance increases over time in 3xTg-AD mice.

Using a Random Forest machine learning regressors we can accurately predict the week that each sample comes from given only its microbiome composition (r-squared: 0.80; p=1.744259e-76, Figure 1). We observed a temporal increase in *Bacteroides* in 3xTg-AD mice beginning at 30 weeks and persisting through 52 weeks. Other taxa, such as *Lactobacillus* did not change due to age or strain.

APP-Wt, APP-Tg, and Tau (JPNL3) mice demonstrated altered microbiome composition in the GI tract. Most notably, the gut microbiota of APP-Wt mice, which are transgenic with the wild-type allele, were distinct from APP-tg mice (Figure 2). Interestingly, different members of the Bacteroidales family seem to drive differences in community composition,

where taxonomy labels and arrows point toward the microbial taxa that are dominant in each mouse type in Figure 2a. Interestingly, despite being on a different genetic background, mice exhibiting tauopathy (JPNL3) appear more similar in microbiota composition to APP-Tg mice than to the wild-type mice exhibiting no Alzheimer’s pathologies. It is important to note that our sample size, in terms of individuals and timepoints, was low in this initial analysis, and that the initial mice available from the vendor were different ages. We have continued to collect these samples for inclusion in an upcoming sequencing run, at which point we will have increased statistical power for assessing this pattern.

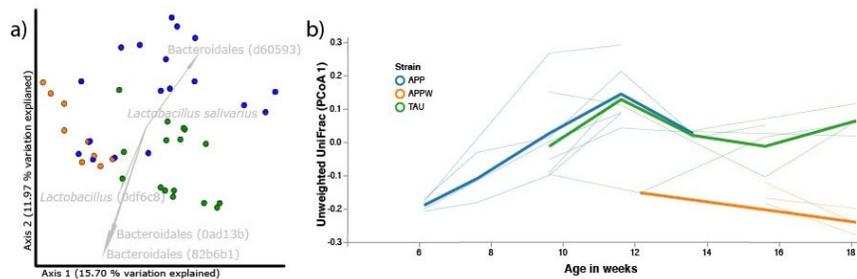


Figure 2: Gut microbiome of AD mouse models are more similar to each other than either is to wild-type mice, suggesting possible AD-related similarities. a) Single transgenic and wild type mice cluster largely independently of one another in an unweighted UniFrac PCoA plot, suggesting differences in their microbiome composition. b) These differences are apparent when tracking changes across mice.

Finally, to assess whether the gut microbiota are driving changes in disease presentation or severity, we used a fecal microbiota transplant (FMT) model. Aged 3xTg-AD mice (12 months) were used as donors. Young 3xTg-AD and wild-type mice received fecal transplants via oral gavage for 5 days immediately post weaning, then fortnightly for 6 months. Mice were sacrificed and gut microbiome, neuroinflammation, and amyloid-beta deposition was assessed. Although we observed a slight increase in IL5 and decrease in ccl2 in 3xTg-AD mice receiving FMT, this was not statistically significant ( $p > 0.05$ , Mann Whitney). The gut microbiota of wild-type mice receiving FMT had more similar GI microbiota to 3xTg-AD mice, indicating at least partial engraftment of the AD-associated gut microbiota (Figure 3c). We are currently quantifying gene expression for other microglial and astrocyte markers in the hippocampus as well as GI inflammation in these mice. Transcriptomics and amyloid-beta immunohistochemistry are currently underway.

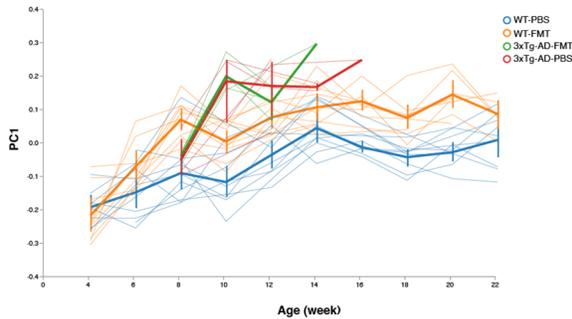


Figure 3. Gut microbiome composition after FMT from aged 3xTg-AD mice. Gut microbiota of wild-type mice transplanted with aged 3xTg-AD shifts toward the 3xTg-AD composition (c).

**Project Progress Reports**  
**Translational Genomics Research Institute**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Correlation between cognitive and motor tasks in an internet-recruited cohort of individuals that span the aging spectrum.** Matt Huentelman, PhD, Lee Ryan, PhD, Sydney Schaefer, PhD. Translational Genomics Research Institute; University of Arizona; Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aim:**

**Investigate the demographic, medical, and lifestyle variables associated with differences in motor and cognitive performance in an internet-recruited cohort of individuals.** Individuals (n=1,000) who participate in our web-based cognitive study, MindCrowd (co-developed by Drs. Huentelman and Ryan), will be invited to complete a motor task that is meant to simulate activities associated with feeding. This motor task was developed by Dr. Schaefer and is simple enough to be performed outside of a laboratory setting. We have devised a version of the task that can be mailed to each study participant and they will perform the task and track their own performance. The results will be returned to the laboratory and we will perform statistical analysis to identify factors associated with motor task performance as well as examine the correlation between the motor and cognitive tasks.

### **Background and Significance:**

Our web-based study of cognition – MindCrowd – has dramatically expanded the size and scope of our knowledge of factors that influence cognitive performance in individuals without dementia across the aging spectrum. However, MindCrowd currently doesn't test motor function in these individuals. The correlation between cognitive and motor function is of great interest to the field as each approach measures different neurological-based functions and therefore allows us to explore the factors that may influence each domain together or separately. By reducing to practice a motor task that is sensitive to aging but also able to be self-administered we are now able to greatly expand the utility of this task and administer it remotely to a large already engaged cohort.

### **Preliminary Data, Experimental Design, and Methods:**

MindCrowd has amassed a database of over 40,000 active e-mail addresses and each individual has consented for future contact regarding other research studies such as the one proposed. The motor task has been developed in such a way as to facilitate self-administration and scoring of the task and the IRB applications are under submission. The motor task has already demonstrated associations with age and other demographic factors in a pilot-sized study group. Our overall goal is to greatly expand the numbers and diversity of individuals who have completed this task by leveraging the MindCrowd resource.

Individuals will be recruited and consented electronically using MindCrowd's database of e-mails. Once consented they will be mailed a package including all of the materials necessary to perform the motor task and record their performance. Data will be either sent back to the lab via mail or entered electronically by the participant. Statistical association analyses will be performed using the demographic, lifestyle, and medical variables collected already via MindCrowd. Additionally survey-style questions will also be administered including a brief history of motor related diseases for each participant.

### **Proposed One-Year and Long-Term Outcomes:**

By the end of this funding year we expect to have motor task data from 1,000 additional individuals from MindCrowd. We will identify factors – medical, lifestyle, and demographic – that will be

associated with statistically different task performance as well as relate motor and cognitive results in the same individuals. Execution of this study will serve as large-scale proof of principle data demonstrating an additional utility of the MindCrowd cohort and the investigators plan to submit federal and foundation grants to expand on this work. Lastly, we expect to draft a manuscript and scientific meeting abstract based on these results.

**Year End Progress Summary:**

We have completed the development of the task for self-administration and scoring. This included the creation of a branded task “game board”, instructions for the test taker, and electronic scoring approach. An instructional video was also produced to guide each test taker on the set-up of the task and game play. We have identified over 5,000 MindCrowd participants for recruitment in our study – this is 5X more than we hope to eventually recruit. We also submitted an NIH R03 grant related to this work. In the remaining Quarter of funding we will test and analyze our goal of 1,000 participants using the self-administered motor task.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Identification of RNA modifications altered in Alzheimer's disease.** Kendall Van Keuren-Jensen, PhD. Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

Chemical modifications are critical to normal cellular functions, however, because of the complexity of modifications and the difficulty in studying them, our knowledge about them is still very basic. It is likely that there are RNA modifications that contribute to Alzheimer's disease pathology, yet this area is almost completely unexplored. According to RMBase, there are more than 100 types of RNA modifications, however, almost nothing is known about how they are regulated, or their function in health and disease (Sun et al, 2015). While the number of protein coding genes has remained relatively stable, the number of non-coding RNAs continues to grow each year. These newly detected RNAs highlight the complexity of the transcriptome and its ability to modulate, stabilize, and change cell functions. The brain is full of exquisite cell types that are unique and carry out exclusive roles. Cells near one another that appear to be similar cell types, have different functions and electrophysiological properties. Single cell sequencing has revealed new subtypes of neurons and glia (Usoskin et al., 2014; Zeisel et al., 2015, Darmanis et al., 2015). If the number of protein coding genes is relatively small, this diversity and regulation of neuronal and glial subtypes must come about through dynamic genetic tools, including the diversity of noncoding RNAs and RNA modifications.

It is well known that some types of RNAs are highly modified, such as tRNAs (Chan et al., 2016), rRNAs (Decatur et al., 2002), and snoRNAs (Meier 2016). There has also been an increasing interest in less abundant RNA modifications on mRNAs (Helm et al., 2017). With increasing advances in sequencing technologies, researchers are finding new ways to study RNA modifications. In this proposal, we want to establish competencies in studying RNA modifications, and use these new tools to reveal important information about AD pathology.

### **Specific Aim:**

We propose to establish new tools and methods with which to examine RNA modifications related to the normal healthy subjects and patients with Alzheimer's disease

One of the main reasons that so few RNA modifications have been identified or studied, has been due to technical challenges. First of all, sequencing, qRT-PCR and most common methods for studying RNA require reverse transcription and conversion to cDNA. These processes strip the RNA of the RNA modifications. Also, RNA modifications often cause reverse transcriptase to fall off of the RNA molecule, causing shortened cDNAs for sequencing. Several papers in recent years have developed strategies to remove the modifications so that the samples can be sequenced and compared (Cozen et al., 2015; Zheng et al., 2015). These new techniques make use of enzymes that can cleave off the RNA modifications. Samples can then be sequenced with and without the enzymes, based on stops in the sequence, it can reveal the location of an RNA modification. We have received the plasmids to grow and purify the enzymes that remove the RNA modifications (Lambowitz). We will use RNA isolated from several different tissues and biofluids. We will also order pre-made RNAs with modifications in locations specified for us, these will allow us to use our analytical tools to try and detect them.

The approach to identification of RNA modifications is direct RNA sequencing. While very few laboratories currently do this, there are early access protocols available using the Oxford Minlon.

This type of sequencer uses a nanopore, rather than the fluorescence and base building chemistry of Illumina. RNAs and DNAs can go through the nanopore which has an ionic current. Each base (G, A, T, C) has a specific disruption on the current and can be read. More importantly, each modification has a specific deflection of current that can also be read. We will take some of the tRNA information we study in the first part of this aim, with known modifications at specific sites, and we will train our ability to assess modifications passing through the nanopore. Oxford nanopore also has a direct RNA sequencing protocol that is in the early stages of testing and was just released. We will adapt this protocol and learn to sequence the RNA directly.

Throughout our tests, we will use cortex from patients with AD, PD and normal healthy controls. We will also use RNAs isolated from biofluids collected from AD, PD patients and normal healthy control subjects. This project is high risk, high reward. If we are able to establish these new tools, and acquire good quality data, we have early access to studying the relationship of RNA modifications and disease.

**Year End Progress Summary:**

First we did preliminary testing between two long read platforms – PacBio and Oxford Nanopore Technologies (ONT). In our hands, ONT did much better, detecting several times the number of transcripts as PacBio. In order to get the most out of the flowcells and samples, we ran pilot experiments and analyzed the data testing several different sample preparation protocols, prior to beginning the proposed experiments. Now that we have established methods (**figures below**), we will proceed.

**Table 1 – Kits Tested**

**\* Compared PacBio and ONT**

**PacBio Tested –**

2 hour pre-extension and 10 hour movie

2 hour pre-extension 20 hour movie

4 hour pre-extension 20 hour movie

**ONT Tested –**

Direct RNA poly A pull down

Direct RNA w rRNA depletion

PCR cDNA

Direct cDNA

**Tested Size selection –**

all transcripts and transcripts > 4000

Figure 1

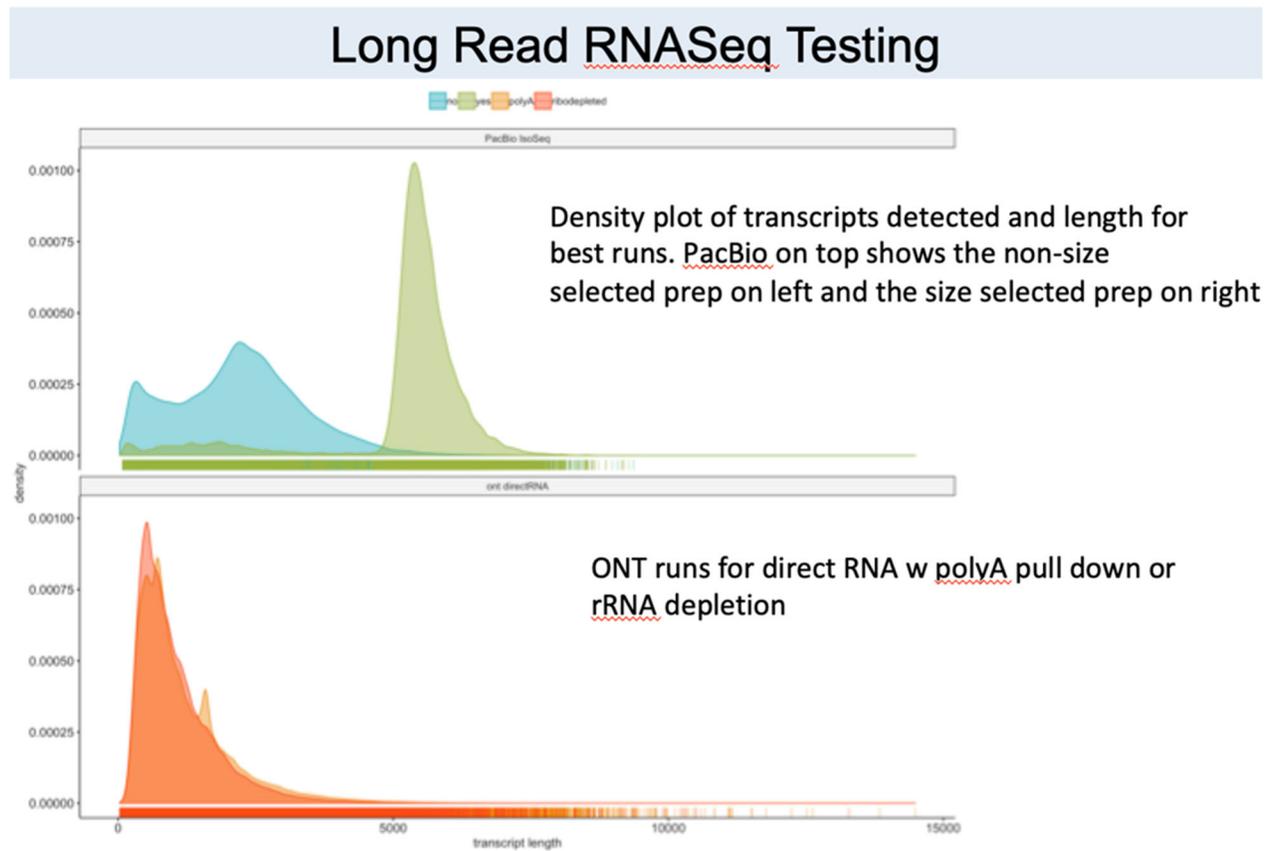
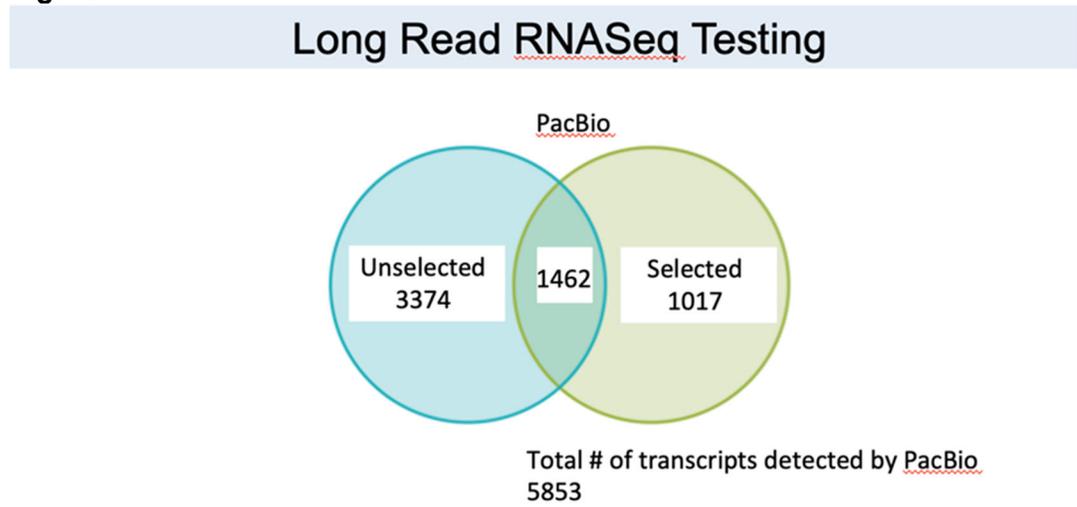


Figure 2



With ONT – we detected - 125,993 detected transcripts, 55,590 > 10 reads

- 1) ONT significantly outperformed PacBio
- 2) We have tested several direct RNASeq approaches – in the next table, and we have found this data will work well

Figure 3

transcriptID	transcript Length	polyA pull down- Direct RNA	polyA + RiboDepletion- Direct RNA	poly A - cDNA
ENST00000387347.2	1,559	67,371	32	304,620
ENST00000418539.2	200	201	252	232,409
ENST00000368865.9	7,703	85	104	82,673
ENST00000361624.2	1,542	43,466	4,798	82,659
ENST00000414273.1	1,543	36,814	4,278	73,635
ERCC-00074	NA	NA	NA	56,309
ENST00000535381.5	5,680	5	10	53,421
ENST00000389680.2	954	21,131	5	51,240
ENST00000361390.2	956	18,109	578	47,720
ENST00000361739.1	684	17,893	1,605	47,290
ENST00000361381.2	1,378	16,731	1,116	47,166
ENST00000362079.2	784	29,159	3,767	42,599
ENST00000361453.3	1,042	13,216	690	41,722
ENST00000361899.2	681	22,455	942	41,242
ENST00000301071.12	1,672	1,529	1,476	39,277
ENST00000342503.8	1,703	994	665	37,008
ENST00000395090.5	1,609	990	665	36,996
ENST00000387342.1	69	290	3	36,965
ENST00000267484.10	3,239	1,033	705	36,950
ENST00000432103.6	2,101	951	669	36,754
ENST00000474911.5	1,412	900	626	36,239
ENST00000514057.1	681	19,816	846	35,467
ENST00000481205.5	3,169	442	406	34,648
ENST00000361789.2	1,141	11,214	1,140	31,176
ENST00000393892.8	5,172	767	600	31,091
ENST00000393888.1	2,583	661	514	30,590
SIRV6	NA	NA	NA	28,866
ENST00000394934.4	2,830	1,156	985	28,327
ENST00000394936.8	2,748	1,157	985	28,319
ENST00000238831.9	11,955	64	77	27,707
ENST00000610929.3	1,969	698	711	27,586
ENST00000600213.3	1,049	4,891	6	24,158
ENST00000387416.2	69	3,977	269	23,456
ENST00000416931.1	372	2,452	78	23,061
ENST00000356978.9	4,203	1,915	1,986	22,824
ENST00000229239.10	1,285	2,870	2,299	22,224
ENST00000396861.5	1,348	2,831	2,286	21,813

The columns under the green headings show the direct RNA compared with cDNA on the ONT platform. Direct RNA sequencing through the pores is slower than how DNA travels through the pores. You can see we detect long and abundant transcript highlighted in yellow

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Cell-specific transcriptomic characterization of the superior frontal gyrus in Alzheimer's disease.** Winnie S. Liang, PhD, Daniel Enriquez, BS, Amir Elyderani, BS, Benjamin Readhead, PhD, Geidy Serrano, PhD, Thomas Beach, PhD, Joel Dudley, PhD, Diego Mastroeni, PhD, Ignazio Piras, PhD, Matthew Huentelman, PhD, Eric Reiman, PhD. Translational Genomics Research Institute; Arizona State University; Banner Sun Health Research Institute; Icahn School of Medicine at Mount Sinai, New York; Banner Alzheimer's Institute; University of Arizona; Arizona Alzheimer's Consortium.

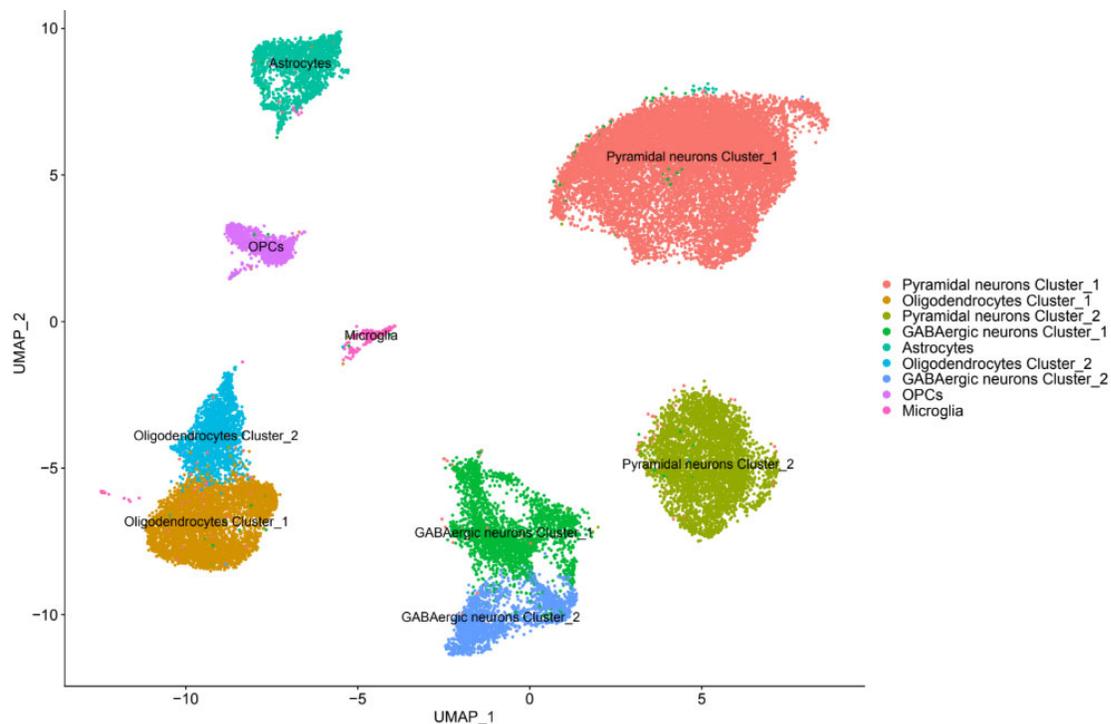
### **Background and Significance:**

We have an ongoing study funded by the Nomis Foundation to generate a region- and cell-specific transcriptomic characterization of brain regions differentially impacted by Alzheimer's disease (AD) for the scientific community. The primary analysis for this proposal is construction of a transcriptomic reference data set generated using laser capture microdissection (LCM) of separate brain cell populations and RNA sequencing (RNAseq). We are complementing this approach using single cell and nuclei strategies (sc/snRNAseq) for characterization and assessment of cell sub-populations.

### **Year-End Progress Summary:**

We have tested, optimized, and evaluated single whole cell, as well as single nuclei, approaches on the 10x Genomics platform. From our comparison of whole cell versus nuclei analyses, we observed that whole cell approaches did not perform as well as single nuclei dissociation, likely due to sample quality as specimens were fresh frozen. This observation was reflected by low expression of genes identified in the AD sample; such expression was also lower in the AD sample

### **Figure 1**



compared to the ND to suggest that the AD sample was more sensitive to dissociation compared to the ND sample. For both the ND and AD preparations, the 5' mRNA approach demonstrated better performance as reflected by a higher number of genes detected per cell. While snRNAseq appears to be more feasible when analyzing fresh frozen brain, we were able to identify distinct cell populations in both the ND and AD brains. We next performed snRNAseq on the superior frontal gyrus (SFG) from 46 ND, MCI, and AD subjects. Identified cell populations include astrocytes, neurons (pyramidal, GABAergic), and oligodendrocytes (see figure).

**Future Directions:**

We have retained leftover single nuclei suspensions for the 46 samples and our next steps are to perform RNA extractions and RNAseq from these suspensions to assess if cell populations can be deconvoluted. We plan to complete sequencing by the summer of 2020 and will complement these analyses with bulk SFG RNAseq and existing snRNAseq data. Additionally, LCMing of astrocytes and microglia is currently ongoing and RNAseq will be performed between 2020 and 2021. Data will be integrated with LCM neuron RNAseq data to build a cell-specific transcriptomic network for the SFG in AD.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Bulk transcriptomic analysis of differentially impacted brain regions in Alzheimer's disease.** Winnie S. Liang, PhD, Benjamin Readhead, PhD, Geidy Serrano, PhD, Thomas Beach, PhD, Joel Dudley, PhD, Diego Mastroeni, PhD, Ignazio Piras, PhD, Matthew Huentelman, PhD, Eric Reiman, PhD. Translational Genomics Research Institute; Arizona State University; Banner Sun Health Research Institute; Icahn School of Medicine at Mount Sinai, New York; Banner Alzheimer's Institute; University of Arizona; Arizona Alzheimer's Consortium.

### **Background and Significance:**

We have an ongoing study funded by the Nomis Foundation to generate a region- and cell-specific transcriptomic Alzheimer's disease (AD) reference for the scientific community. We are constructing this reference using complementary strategies of which the foundation is laser capture microdissection and RNA sequencing (RNAseq) of specific cell populations across differentially impacted brain regions from healthy elderly control and AD subjects. The goal for the study here is to generate bulk RNAseq data from the superior frontal gyrus (SFG), the same brain region for which we have completed non-tangle-bearing neuron collections and RNAseq, as well as single nuclei RNAseq (snRNAseq) on the 10x Chromium platform. The purpose of performing bulk RNAseq is to assess if cell populations identified using LCM RNAseq, and/or snRNAseq, can be deconvoluted from bulk RNAseq data. As the majority of publicly-available AD and brain RNAseq data is not cell-specific, development and optimization of a approach to infer cell populations will be valuable for interpreting and analyzing existing data, and will also enable future studies as cell-specific transcriptomic analyses is a timely and costly process.

### **Year-End Progress Summary:**

The originally proposed project for this study was a collaborative study with the Mayo Clinic in Arizona to assess the impact of an intervention on the gut microbiome of mild cognitive impairment (MCI) subjects. It was not determined until February 2020 that this project could not proceed due to administrative and financial reasons. As a result, we are proceeding with the bulk RNAseq study described here. We have begun optimizing a total RNA extraction protocol on the PerkinElmer chemagic 360 instrument to enable automation of extractions which will aid in the reduction of batch effects from sample processing. We have also received fresh frozen SFG sections from the Brain and Body Donation Program (BBDP) at the Banner Sun Health Research Institute, and plan to complete total RNA extractions and stranded total RNAseq from approximately 50 healthy elderly control and AD subjects in the spring of 2020. All libraries will be paired-end sequenced for 100bp reads on the Illumina NovaSeq 6000 to target 150 million total reads per sample. Data will be processed and analyzed through TGen's internal Jetstream and Phoenix pipelines. Bulk RNAseq data will be analyzed in collaboration with Dr's Piras and Huentelman who are developing transcriptomic cell deconvolution strategies.

### **Future Directions:**

We will further expand bulk RNAseq analyses to additional brain regions we are concurrently analyzing through our Nomis-funded study on the same set of approximately 50 subjects. These regions include the entorhinal cortex, hippocampus, posterior cingulate, and primary visual cortex.

**Project Progress Reports**  
**University of Arizona**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Influence of Physical Activity and Sleep Quality on Brain Aging and the Risk for Alzheimer's Disease.** Gene Alexander, PhD, David Raichlen, PhD, Geoff Ahern, MD, PhD, Thomas Beach, MD, PhD, Richard Caselli, MD, Yi Su, PhD, Matt Huentelman, PhD, Yann Klimentidis, PhD, Eric Reiman, MD, Ted Trouard, PhD, Lee Ryan, PhD. University of Arizona; Banner Sun Health Research Institute; Mayo Clinic; Banner Alzheimer's Institute; Translational Genomics Research Institute; Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

This highly integrative and collaborative research project will leverage expertise across the state of Arizona with the Arizona Brain Body Donation Program (BBDP), the Arizona Alzheimer's Disease Core Center (ADCC) Brain Imaging and Fluid Biomarkers (BI-FB) Core, and the University of Arizona to support implementation of physical activity (PA) and sleep quality measures to investigate how health and lifestyle factors interact with cerebrovascular risk, brain gray and white matter, and Alzheimer's disease (AD) brain pathology to influence cognitive aging and preclinical AD risk. The following specific aims will be addressed: 1) to determine how PA and sleep quality influence cognitive performance in older adults with differential risk for AD; and 2) to develop, evaluate, and implement novel methods for processing and analysis of PA and sleep quality time-series data to identify new biomarkers for age-related cognitive decline and AD risk.

Additionally, this study provides added value with key pilot data to: 1) evaluate wearable technologies as novel biomarkers for the Arizona ADCC BI-FB Core, 2) create a unique dataset to support cognitive aging and AD research across Arizona, 3) explore how brain imaging and genetics relate to PA and sleep in older adults; 4) evaluate how PA and sleep ultimately relate to post-mortem brain pathology, and 6) support new external grant proposals on aging and AD risk.

### **Background and Significance:**

The population of older adults is expected to grow rapidly over the next two decades and it will be important to respond to the associated growth in AD, in Arizona and nationally. Whereas genetic (e.g., APOE e4) and cerebrovascular risks increase AD risk, exercise can improve cognition in aging and may reduce AD risk, yet the mechanisms underlying these benefits have not been fully elucidated. In older adults, high PA levels are associated with greater brain volume and connectivity. Studies of cognition and brain imaging in older adults are critically needed to identify how PA supports healthy brain aging, while reducing AD risk. Sleep quality is another important aspect of daily activity that may impact brain aging and AD risk. This proposal requests support for a cross-institutional, highly collaborative research project, including investigators from the University of Arizona, Banner Sun Health Research Institute, TGen, and Banner Alzheimer's Institute. Our overarching hypothesis is that PA and sleep quality moderate the impact of cerebrovascular, genetic, and other health factors to influence brain aging and AD risk by altering brain networks important for cognitive processes that depend on frontal and temporal brain regions and the integrity of connecting white matter. The project will provide the essential data and methodological developments in support of a larger multi-disciplinary research program with the goal of advancing our understanding of how differences in daily PA impact brain aging and the risk for AD. We have established a collaborative team of Arizona Alzheimer's Consortium (AAC) investigators, with expertise in neuropsychology, neurology, neuroimaging, genetics, statistics, biomedical engineering, and exercise/sleep behavior. The proposed research will

implement “state-of-the-art” PA and sleep quality measures using actigraphy and self-report scales to augment tests of cognition, MRI brain scans, PET imaging of AD pathology, CSF/blood biomarkers, genetics, and post-mortem brain pathology already implemented for a unique Arizona older adult cognitively-unimpaired cohort, as part of the Arizona BBDP and BI-FB Core.

### **Year End Progress Summary:**

We have made significant progress in our studies on health factors and lifestyle characteristics for brain aging and the risk for AD. Data collection for physical activity and self-report measures in the unique Arizona cohort of the Brain Body Donation Program in a collaboration between the University of Arizona, Banner Sun Health Research Institute, TGen, and the Banner Alzheimer’s Institute, is well underway, providing a valuable resource for AD and aging research for use by multiple investigators at the University of Arizona, as well as across Arizona and nationally. In this project, we have demonstrated the feasibility of acquiring high quality actigraphy and self-report data on physical activity, as well as collection of blood spot samples from healthy community dwelling older adults in Arizona. It is expected that continuing to build this dataset will provide a key cohort to support the submission of future collaborative grant proposals by Arizona investigators, and we have already begun to leverage this collaborative effort to support new grant submissions and our plans for our upcoming NIA center grant renewal. A manuscript applying our novel method of actigraphy pattern analysis has been published showing that less consistent physical activity was associated with increasing age and greater mortality (Raichlen et al., *Journal of Gerontology: Biological Sciences and Medical Sciences*, 2019). In addition, we were invited to publish an article in *Scientific American* highlighting our evolutionary-neuroscience model of physical activity and brain aging (Raichlen and Alexander, *Scientific American*, 2020), which was featured on the cover of the January 2020 issue. We have two publications showing how physical activity is both related to brain structure (Raichlen et al., *Brain Imaging and Behavior*, 2019), and can enhance cognitive function during dual-task walking (Raichlen et al., *BMC Geriatrics*, in press). An article was published demonstrating spatial learning deficits in a novel transgenic rodent model of hypertension (Willeman et al., 2019). We also have articles submitted for publication investigating the potential impact of pollution on brain structure in an aging cohort (Furlong et al., submitted), evaluating the effects of aging and physical frailty in a novel cross-cultural cohort of East African foragers and pastoralists (Sayre et al., submitted), and evaluating how differences in hypertension status influence the mediation of subjective memory complaints by hippocampal atrophy in healthy aging (Van Etten et al., submitted).

Our \$3.8M NIA grant to supplement our NIA Arizona Alzheimer’s Disease Center (ADC) has established a collaborative Brain Imaging and Fluid Biomarkers Core (Core Leader: Alexander; Co-Investigators: Reiman (ADC PI), Beach, Ahern, Kuo, Trouard, Ryan, Su), providing enhanced access and expertise for the use of MRI, PET, CSF, and blood biomarkers in collaborative AD and aging research across Arizona. We plan to further incorporate our physical activity lifestyle measures as additional technology-based biomarkers into this core to provide expanded cutting-edge markers for preclinical AD risk and to support our plans for our NIA core center grant renewal. In the past year, a new \$3.8M collaborative NIA grant has been awarded to the University of Arizona and University of Florida, Gainesville to evaluate the use of near-infrared stimulation to reduce the risk of AD (MPIs: Alexander, Bowers, Woods). A new \$120K collaborative pilot study between the University of Arizona and University of Florida, Gainesville has been funded by the McKnight Brain Research Foundation to test a novel approach to reduce brain aging using vagus nerve stimulation (PI: Williamson, Site PI: Alexander). A new \$1.7M NIA grant was awarded to the University of Arizona to evaluate the neuroimaging and cognitive effects of a novel cognitive decision-making task in older adults (PI: Wilson; Co-Investigators: Alexander, Ekstrom, Chou, Andrews-Hanna). An application for a \$3.6M NIA grant was submitted by University of Arizona investigators to study enhancement of hippocampal plasticity using transcranial magnetic

stimulation in those at risk for AD dementia (PI: Chou; Co-Investigators: Alexander, Barnes, Bedrick, Chen, Fisher, Mohler, Rapcsak, Ryan). Additionally, a new \$3.8M NIA grant was submitted from the University of Arizona to investigate how physical activity influences brain aging and the risk for AD (MPIs: Alexander, Raichlen) and a new \$300K NIA supplement grant was submitted to develop methods for measuring cognition and physical activity in a novel animal model of age-related cognitive decline and dementia (PI: MacLean; Co-Investigator: Alexander).

Work from this AAC project also supports the development of new methods and complements ongoing studies of physical activity and sleep quality assessment of healthy oldest old adults funded by the McKnight Brain Research Foundation (MPIs: Alexander, Cohen, Visscher, Rundek) to evaluate how lifestyle factors influence cognition and brain aging in older adults, ages 85 to 100+. This complementary effort is currently underway and reflects ongoing collaborations between the University of Arizona, University of Florida, University of Alabama, and the University of Miami. Initial findings from this work have shown that, among oldest old adults, engaging in more moderate to vigorous activity is associated with greater brain volume in regions of frontal cortex. An abstract of this work was presented at the Society for Neuroscience (SFN) in Chicago, IL in October 2019.

In additional abstracts presented at the 2019 SFN meeting in Chicago, IL and the 2020 International Neuropsychological Society (INS) meeting in Denver, CO, we also showed 1) that APOE  $\epsilon$ 4 status influences the mediation of the relation between aging and hippocampal volume by temporal lobe white matter hyperintensity lesion volumes (Van Etten et al., SFN, October, 2019), 2) that expression of Alzheimer's disease biomarker-related gray matter covariance patterns differ in those with and without subjective cognitive decline (Bharadwaj et al., SFN, November, 2018), 3) that the interactive effects of white matter hyperintensities and gender on heart rate response to exercise may reflect a novel behavioral biomarker in aging (Song et al., INS, 2020), 4) that age-related covariance patterns of cortical thickness are related to cognitive aging (Smith et al., INS, 2020), and 5) that white matter integrity mediates the relation between body mass index and executive function in older adults (Van Etten et al., INS, 2020).

In addition, Dr. Alexander was invited to present findings from his work on physical activity, brain aging, and the risk for AD at Loma Linda University Medical Center in Loma Linda, CA, at the McKnight Brain Research Foundation Inter-Institutional Meeting in Miami, FL, and at the Fellows meeting of the American Psychological Association Adult Development and Aging, Division 20 in Washington, D.C.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Real-world markers and neural mechanisms of Alzheimer's disease risk in cognitively normal older adults.** Jessica R. Andrews-Hanna, PhD, Matthew D. Grilli, PhD, Matthew Huentelman, PhD, Matthias Mehl, Lee Ryan, PhD. University of Arizona; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

This project seeks to uncover sensitive real-world predictors and neurocognitive mechanisms underlying two major risk factors for Alzheimer's disease (AD): depression and presence of the APOE4 allele. Leveraging our development of two accessible mobile smartphone apps, we will test the hypothesis that depressive symptoms and APOE4 alter thought and social interaction in daily life in different ways, and increase AD risk through disparate neural mechanisms. The proposed pilot studies will best position our team, and others in the Arizona Alzheimer's Consortium (AAC), to apply for federal funding to effectively measure, monitor, and impede AD risk, and ensure that one's later years of life remain as healthy and as productive as possible.

### **Specific Aims:**

- 1) To test the hypothesis that two established risk factors for AD – depressive symptoms and APOE4 genetic status – are differentially linked to alterations in everyday thought and social conversation in a seemingly cognitively normal (i.e., no mild cognitive impairment or dementia) older adult cohort.
- 2) To test the hypothesis that subjective and objective naturalistic measures of thought and social dialogue predict daily functioning and social well-being better than standard laboratory-based tests.
- 3) To test the hypothesis that depressive symptoms and APOE4 may impede daily functioning and social well-being through disparate functional network connectivity aberrations within the brain's default network (DN).

### **Background and Significance:**

Older adults represent the fastest growing segment of the population, igniting a particular urgency for researchers and healthcare professionals to ensure that these later years remain as healthy and as productive as possible. Although a large body of work investigating cognitive changes in normal and pathological aging has established domains of cognition especially vulnerable or resistant to cognitive decline, the vast majority of evidence comes from controlled experimental paradigms conducted in laboratory settings that lack the varied environmental contexts and cognitive demands of everyday life. To fill this gap, the proposed project will establish a *Naturalistic Assessment Core (NAC)* as part of a multi-disciplinary, collaborative aging project among members of the AAC. By leveraging our team's development of two mobile smartphone apps, combined with our expertise in functional MRI network analyses and neuropsychology, this pilot funding proposal will allow us to develop more sensitive, cost-efficient, and accessible neurocognitive markers of AD-related risk that can be monitored remotely over time. These biomarkers may provide clinicians and patients with unique insight into mechanisms of risk and targets of intervention, contributing substantially to efforts to improve quality of life in later years.

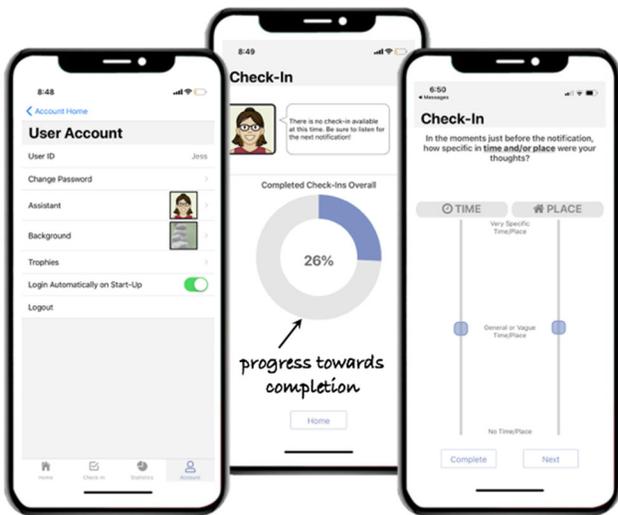
### **Year End Progress Summary:**

- 1) Data collection for our proposed ACHIEVE study is ongoing; as planned, we are optimistic about meeting our proposed sample size by the end of the funding cycle.

2) This past year, PI Andrews-Hanna's team has successfully developed, branded and piloted a free cross-platform mobile research app, called Mind Window, that we believe will have important implications for how the aging field understands healthy and pathological aging. We are using Mind Window in the context of the *ACHIEVE* study's *Naturalistic Assessment Core* to assess internal fluctuations in mood,



To make the experience more engaging, users can select a "check-in assistant," choose a display background, and win trophies for different stages of completion.



memory, and thought content as individuals go about their day-to-day lives. We are also employing our team's 2<sup>nd</sup> app, called the Electronically Activated Recorder, as a means to concurrently assess "externalized" experience that our participants share in social conversation.

- 3) Within the last year, Dr. Andrews-Hanna and Dr. Grilli submitted (as MPIs) a \$3,088,000 R01 to NIH/NIA titled "Tracking autobiographical thoughts: a smartphone-based approach to the detection of cognitive and neural markers of Alzheimer's disease risk." This is a collaborative grant that also includes Co-Is Huentelman and Mehl, and would not have been possible without the AAC pilot funding that generated critical preliminary data. The grant received promising scores 5% away from the funding line, and we are now in discussion with the Program Officer about next steps.
- 4) Using AAC pilot data and the Mind Window app, Dr. Andrews-Hanna prepared another R01 grant proposal (as MPI) for a March 2020 submission (PAR-13-373) investigating mechanisms underlying links between depression and social connectedness in young adults and midlife (\$3,247,698).
- 5) Through the AAC and prior pilot funding, Dr. Andrews-Hanna helped prepare a recently-awarded NIH/NIA R01 grant as Co-I (PI = AAC member, Robert Wilson) to examine neurocomputational mechanisms of decision making in older adults. Prior AAC funding awarded to Dr. Robert Wilson and Dr. Ying-hui Chou facilitated data collection and kick-started this collaboration that led to a successful 5-year federal grant.
- 6) In accordance with the overarching goal of this project, Drs. Grilli, Andrews-Hanna, and Mehl contributed to a study examining the naturalistic assessment of autobiographical memory and future thinking in older adults (Wank et al., under review), which was invited to be part of a special issue in *Frontiers in Human Neuroscience*.
- 7) Dr. Andrews-Hanna and Dr. Grilli are currently preparing an invited theory piece for a high impact journal: *Current Directions in Psychological Science* that includes a discussion of naturalistic assessment of cognition (first draft deadline 4/15/2020).

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Role of medin membrane pores in AD and vascular-dementia-related vasculopathy.**  
Fernando Teran Arce, PhD, Raymond Migrino, MD, Jill Madine, PhD. University of Arizona; Phoenix VA Health Care System, University of Arizona College of Medicine-Phoenix; University of Liverpool, UK; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1. Electrophysiological activity of medin pores using model membranes.
2. Morphology of membrane-associated, pore-forming, medin species.
3. Link medin EC cytotoxicity to pore-forming ability.

### **Background and Significance:**

Medin is a common, yet rarely studied, amyloidogenic protein that accumulates in the vasculature with aging. It has 50 amino acids and shares structural features with the A $\beta$  peptide, the putative agent underlying Alzheimer's disease (AD). We previously showed that medin accumulates in the vasculature with aging, and that cerebral arteriole medin is higher in vascular dementia (VaD) than in cognitively normal brain donors. We also showed that physiologic doses of medin induced endothelial dysfunction and EC immune activation, while also causing EC cytotoxicity. We further demonstrated that medin is present in cerebral arterioles, suggesting that it may cause microvascular endothelial dysfunction. We will build on these discoveries to determine if dysregulated ionic flux induced by membrane pores is a mechanism of medin injury. Our previous work suggests that A $\beta$  oligomers form pore structures that permeabilize the membrane, altering cellular homeostasis. We focus on this **membrane-permeability-increasing mechanism** because it is believed that: i) *oligomeric species formed in the early stages of aggregation are cytotoxic*, and ii) toxic processes likely involve the interaction of oligomeric species with cell membranes.

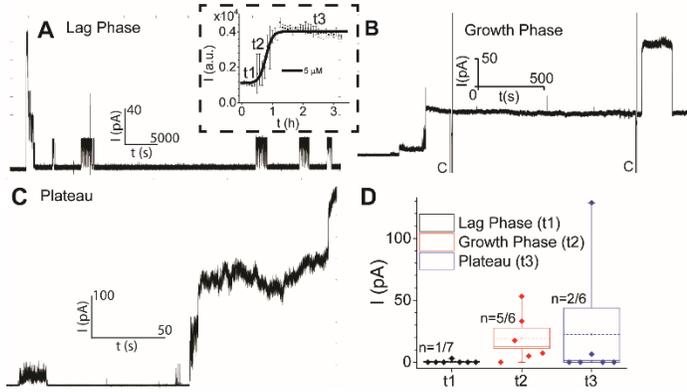
### **Year End Progress Summary:**

**Accomplishments.** A manuscript titled "Medin Oligomer Membrane Pore Formation: A Potential Mechanism of Vascular Dysfunction", based on the work described in Aims 1 and 2 of this report, was submitted to Biophysical Journal. It received very good reviews by the referees and it will likely be accepted for publication upon resubmission.

1. **Goal.** To answer the question: do medin oligomers form membrane pores that lead to ionic permeability of a lipid membrane?

**Methods.** We used planar lipid bilayer (abbreviated BLM for "black lipid membrane") electrophysiology as a simple biophysical model system to test if medin oligomers in well-defined aggregation states lead to membrane pore formation. Membranes were composed of: i) an equimolar mixture of phospholipids used previously to test A $\beta$  pore formation, and ii) a lipid composition closer to membranes of endothelial cells. As sample agitation was critical for medin aggregation, the Thioflavin T (ThT) fluorescence of replicas of the medin samples used for electrophysiology was measured to monitor their aggregation state. Samples in the three stages of amyloid aggregation were used (inset, Fig. 1A); the lag phase (t1), the growth phase (t2), and the plateau phase (t3). Following sample agitation, medin was added to one of the electrolyte compartments separated by the BLM, and the ionic current (I) flowing through the membrane was monitored as a function of time (t) in I vs. t traces.

**Progress.** Using BLM electrophysiology, we found sudden, stepwise increases of ionic current through lipid bilayers upon exposure to oligomeric medin species (Fig. 1, A - C); indicative of the formation of medin membrane pores. Medin pores also displayed burst events, indicating rapid opening and closing of small pores, and spike events, i.e., short-lived step events with durations of a few milliseconds to seconds. I vs. t traces acquired over several hours could exhibit

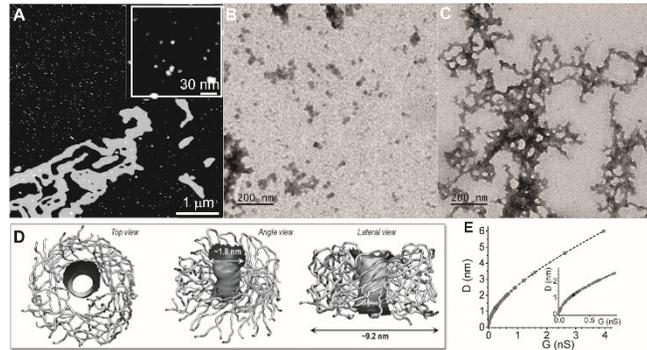


all these different behaviors. The ionic permeability induced by medin proteins in the lag phase was significantly lower than in the growth - and plateau phases (Fig. 1D). More than 80% of medin samples in the growth phase displayed pore activity, compared to the activity of medin samples in the lag and plateau phases (14 % and 33 %, respectively). Growth phase samples consistently induced pore activity and some of the pore events had large conductances, which were stable over tens to thousands of seconds. These high-conductance pores have the potential of significantly altering cellular homeostasis and inducing rapid cytotoxicity by a single event (e.g., pores with conductances above 1 nS could induce lethal cytotoxicity in seconds). The pores formed had a broad distribution of step conductances, with the majority of values between 10 – 1000 pS, which correspond to pore diameters in the range of 0.1 – 2.5 nm. Two pores from growth phase samples had extremely large diameters (4 – 6 nm).

**2. Goal.** To characterize the structure of medin aggregates and pore-forming species.

**Methods.** We used atomic force microscopy (AFM) to image the morphology of medin samples in three stages of their ThT aggregation curve (Fig. 1); 1) before agitation (start of lag phase), 2) agitated until ThT fluorescence was detected (growth phase), and 3) agitated for two hours after the plateau was reached.

**Progress.** Two types of aggregates were discerned in the growth phase; large flat domains with heights of 4.7 nm, occupying areas of several tens of  $\mu\text{m}^2$  (Fig. 2A), and much smaller structures with diameters of  $\sim 20$  nm (left side of Fig. 2A). By performing particle size analysis to estimate the volumes of the oligomers seen in the growth and lag phases, we found each average-sized oligomer is composed of  $\sim 200$  monomers, too large to form pores. Almost all the structures observed in the plateau phase were similar to the large domains seen in the growth phase. Significantly, no fibers could be identified. The domains resemble the morphology of lipid bilayer patches formed by rupture of liposomes on a solid surface. In addition to the domains, samples in the growth- and plateau-phases contained populations of small oligomers with sizes compatible with pore-forming species (inset, Fig. 2A). Transmission electron microscopy (TEM) images of the medin aggregates (Fig. 2, B and C) displayed morphologies which were consistent with the small aggregates seen in the AFM images (Fig. 2B), and additional open structures, which suggest the presence of pre-fibrillar aggregates and were not seen by AFM (Fig. 2C). The lack of fibers in the AFM and TEM images, the planar domains formed by medin aggregates, and



their low ThT fluorescence compared to A $\beta$  fibers, suggests a non-amyloidogenic pathway to pore formation.

We collaborated with Dr. Ruth Nussinov's Computational Biology group at the National Cancer Institute to model the structure of medin pores *in silico*. The results of our all-atom molecular dynamics simulations showed 18-mer pores, with toroidal structures rich in  $\beta$ -sheets ( $\beta$ -barrels), which were stable both in zwitterionic and anionic bilayers (Fig. 2D). Their ionic selectivity was low compared to A $\beta$  pores. The pore diameters were  $\sim 1.8$  nm for the anionic membranes, corresponding to pores with  $\sim 500$  pS conductances, well within the range of the conductances found experimentally (Fig. 2E). Circular dichroism spectroscopy measurements showed that the aggregated species possessed  $\beta$ -sheet structures, consistent with our *in silico* models. Medin samples in the lag phase had several structures with heights below 1 nm, which could only be identified at high magnification imaging. The estimated volumes ( $\sim 9$  nm<sup>3</sup>) are slightly larger than the volume of the U-shaped monomers used in our *in silico* modeling ( $\sim 5.5$  nm<sup>3</sup>), suggesting that these lag-phase structures are primarily composed of medin monomers and low-n oligomers, which are too small to form pores. The multiple conductances of amyloid pores suggest that pores have different diameters and consist of varying numbers of monomers.

**3. Goal.** To determine if medin causes endothelial cell toxicity in part through abnormal formation of membrane pores.

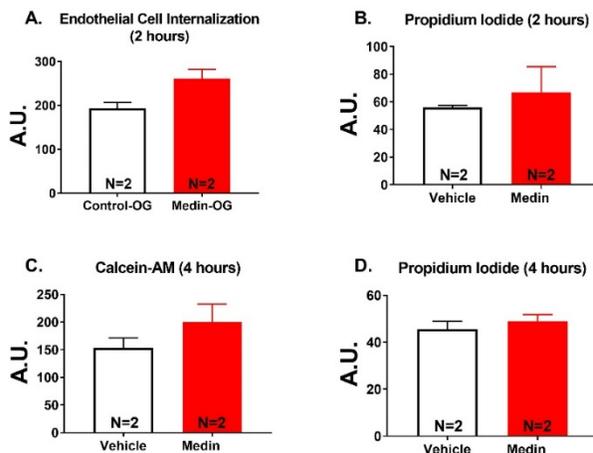
**Methods.** In *Experiment 1*, our goal was to determine if exposure of primary human microvascular endothelial cells (ECs) to medin causes medin internalization and if this process is associated with increased cellular uptake of propidium iodide, PI (uptake of PI indicates membrane disruption). We selected a short incubation time (2 hours) as this was a time that membrane pores could form (leading to medin internalization), yet short enough not to expect cell death processes that could also lead to PI uptake. Here we expect that any PI uptake will be due to membrane disruption that is not due to necrosis.

Recombinant medin conjugated with Oregon green (OG) was used. Control protein (scrambled protein) conjugated with OG was used. Medin-OG was agitated for 1 hour. ECs (N=2) were exposed for 1 hour to medin-OG (5  $\mu$ M) and control-OG and fluorescent signal measured in Biotek. Separately, ECs were exposed to medin or vehicle control (N=2) for 1 hour followed by administration of PI and fluorescent signal was measured.

In *experiment 2*, we increased the exposure time to medin to 4 hours. After 4 hours, we applied PI (to assess membrane leak) and calcein AM (to assess viability). If medin causes membrane pores, we expect PI signal to increase with medin treatment, but because of short exposure time (4 hours), we expect that the cells will remain viable (same calcein AM).

**Results.** There was a trend towards increased signal in Medin-OG versus control-OG for *experiment 1* (Fig. 3, A and B). If confirmed with additional replicates, this would indicate medin internalization by cell. PI signal is shown in B. Interpretation requires a bigger sample size. There is no discernible trend for *experiment 2* (N=2) (Fig. 3, C and D). Larger sample sizes are needed.

**Conclusions:** Technical aspects of planned experiments are working well and we will proceed with increasing sample size after the process of optimization. If PI signal remains low at 2-4 hour exposure of medin, we will confirm with flow cytometry based reading to verify PI.



# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Age-related specific changes in expression of several central melanocortin receptor subtypes and their localization in rat brain.** Carol A. Barnes PhD, Minying Cai PhD, Victor Hruby PhD, Ted Trouard PhD. University of Arizona; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) Conduct a behavioral test battery that specifically interrogates the cognitive function of different cortical regions, and in the same animals, assess specific melanocortin receptor subtype (MCR) binding density in old and young rats.
- 2) Determine the brain localization of a novel MCR-selective ligand (Cai and Hruby). We will accomplish this by using gadolinium enhancement for MRI imaging (7T magnet, Trouard). A time course of brain bioavailability will be determined in this study (predictions range from 1 to 4 days)
- 3) After completion of Aim 2, which will establish brain penetration and distribution of this novel compound, we will treat one group of old rats with the compound and one group with a control solution for a period of several weeks. The frequency of dosing will be determined by the time course of bioavailability established in Aim2. Animals will then be given a series of behavioral tests to assess whether cognition is superior in the treated versus untreated animals.

### **Background and Significance:**

The melanocortin system has been associated with the control of many physiological functions critical for survival, these include feeding behaviors, pigmentation, control of inflammatory disorders, immunomodulation, antipyretic effect and prevention of brainstem ischemia and reperfusion injury (Gonzalez et al., 2009). The melanocortin system consists of five distinct receptors (hMC1-5R) that belong to class A of seven-transmembrane G protein-coupled receptors (GPCRs). It has been reported that the human melanocortin 4 receptor (hMC4R) is involved in neurodegenerative disease (Shen et al., 2013). Melanotropins have also been proposed as agents that may protect against the progression of Alzheimer's disease (Giuliani et al., 2014). Furthermore, administration of  $\alpha$ -MSH or its more stable analog [Nle<sup>4</sup>,D-Phe<sup>7</sup>]- $\alpha$ -MSH (NDP- $\alpha$ -MSH) has been observed to enhance learning and memory in young animals (Beckwith, et al. 1975). Thus, there is some support in the literature for the hypothesis that melanocortin manipulation may improve cognition in aging.

The impact of age with respect to melanocortin receptor expression, however, remains unexplored. Thus, our findings may potentially open a new window of discovery for exploring and developing new treatments for cognitive changes that arise in normal aging and in neurodegenerative diseases.

### **Year End Progress Summary:**

We completed the study of the 5 melanocortin receptor subtypes in young and aged behaviorally characterized rats. Six regions of the brain were extracted from each animal, including the frontal cortex + anterior midbrain, parietal cortex, cerebellum, posterior midbrain, hippocampus and occipital lobe. We collected the membrane fragments from each region of all animals in each age group, then ran a specific binding assay using iodine labelled NDP- $\alpha$ -hMCHR1-5 on a high throughput Micro Beta II radiation counter. Six samples were measured from each animal for each

region, and then averaged to produce a single count for each animal in each region. All measurements were collected in a blind fashion.

We ran a linear regression analysis with spatial learning behavior and specific receptor binding for hMCHR1-5 receptors using Graph-pad Prizm software. A significant correlation was found between spatial memory and two receptor subtypes MCH-1R ( $p = 0.048$ ) and MCH-3R ( $p = 0.049$ ) in old animals. This finding potentially opens a new window of discovery for exploring and developing new treatments for cognitive changes that arise in normative aging and in neurodegenerative disease.

Minying Cai and Carol Barnes have submitted an R21 NIH grant entitled “Study of Melanocortin Receptors Expression Correlated with Spatial Memory” to secure funding to support further experiments using the pilot data collected from this project as supporting evidence.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Neuroinflammation, Aging, and Cognition: An Intervention Study.** Carol A. Barnes PhD, Meredith Hay, PhD, John F. Guzowski, PhD. University of Arizona; University of California Irvine; Arizona Alzheimer's Consortium.

### **Project Description:**

Administer a cognitive behavior battery to assess temporal lobe and frontal lobe functions to assess the impact on cognition of a 2 month minocycline treatment. We will also characterize the neuroinflammatory status of the hippocampus and prefrontal cortex.

### **Specific Aims:**

- 1) To investigate antibiotic intervention therapy on age-related cognitive decline
- 2) To determine the inflammatory and microglial response to chronic minocycline treatment in middle aged rats

### **Background and Significance:**

Aging is associated with a general increase in inflammatory tone in the brain. Elevated neuroinflammatory processes are thought to contribute to cognitive decline in both healthy aging and in neurodegenerative diseases such as Alzheimer's disease. Moreover, increased neuroinflammation is implicated as a primary driver of cognitive impairment in a number of neurodegenerative, neuropsychiatric, and neurodevelopmental disorders. The FDA-approved tetracycline-derived antibiotic minocycline has been shown to have anti-inflammatory actions in brain by inhibiting the activation of microglia to immune challenges and during aging. For this reason, and its widespread availability and low toxicity in patients, minocycline is an attractive drug for combination therapies in which the goal is to attenuate neuroinflammation to restore cognitive function in human patient populations. However, minocycline has other potential neuroprotective actions that may provide additional pro-cognitive benefits. In animal studies, minocycline treatment has been shown to protect memory function against specific immune challenges or restore memory function in aged animals. To date, however, little attention has been paid to determining how minocycline provides these pro-cognitive effects; it is generally assumed that it is through reducing neuroinflammation, without considering other potential neuroprotective effects such as modulation of pathways involved in the generation of reactive oxidative species.

### **Year End Progress Summary:**

Animals arrive in the laboratory at ~15 months of age (which is equivalent to 'middle age' in these rats). They were split into two groups of rats – minocycline-treated (70mg/kg/day), and placebo, for 8 consecutive weeks before the beginning of the behavioral test battery. This ~ 2 month test battery was begun with the rats were roughly 18mo of age, and consisted of spatial and cued watermaze, spontaneous object recognition, hippocampus region-specific cheeseboard testing as well as motor (rotorod) and anxiety tests (EZ maze). There were no behavioral differences detected between treatment and control groups at that time.

There could be a number of factors that contributed to the results observed – two of which were include the possibility that the reduction of inflammation would only be detected after longer treatment intervals, or at older ages. Thus, we continued the treatment for an additional two months, and repeated the cognitive test battery when they were ~20 months of age. We

computed “savings scores” for the tests that would exhibit “test-retest’ effects – but none of the data suggested a cognitive advantage for the treatment group.

Our current thinking is that chronic administration of minocycline may only be therapeutic in the context of greater neural injury and stress than what is seen in normative aging - the degree of neuroinflammation mediated by microglia in the aging brain may be below the threshold necessary to benefit from the anti-inflammatory effects of minocycline. This may explain why in TBI models, for example, minocycline is effective, but in our case it is not.

We are currently writing these data up, as they should provide important information for experiments designed to explore potential neuroprotective effects of minocycline and the contexts in which these beneficial effects may be observed.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Characterization of dynamic CBF and BOLD signals of the hippocampus with amnesic mild cognitive impairment.** Nan-kuei Chen, PhD, Ying-hui Chou, ScD, Manoj Saranathan, PhD, Gloria Guzman Perez-Carrillo, MD, Ashley Stokes PhD. University of Arizona; Barrow Neurological Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

Changes in structure and function of the hippocampus, measured by MRI, have been observed in subjects with Alzheimer's disease (AD) and amnesic mild cognitive impairment (aMCI). However, the association among local MRI signals (e.g., hippocampal CBF), the intrinsic connectivity network integrity (e.g., the functional connectivity between the hippocampus and other default-mode network nodes: measured by both BOLD and dynamic CBF scans), and the behavioral performance (e.g., the memory performance) has not yet been thoroughly studied in aMCI subjects. In this project we propose to develop a robust MRI protocol to simultaneously measure dynamic CBF and BOLD signals (specifically: 80 CBF time points and 80 BOLD time points obtained in an interleaved manner), from which the association between local MRI signals (e.g., hippocampal CBF) and the intrinsic network connectivity can be quantified. We will acquire interleaved CBF-and-BOLD data from 18 aMCI subjects and 18 healthy controls, and then measure the association among local hippocampal measure, functional network integrity and the memory performance across subjects in each of the two populations.

### **Specific Aims:**

- 1) To implement an interleaved arterial-spin labeling (ASL) and T2\*-weighted BOLD MRI pulse sequence that can robustly and simultaneously measure dynamic CBF and BOLD signals
- 2) To implement post-processing algorithms that can effectively suppress non-physiological signal fluctuations in the interleaved CBF-and-BOLD data
- 3) To measure the connectivity between hippocampus and other default mode network nodes from the interleaved CBF-and-BOLD data obtained from subjects with and without aMCI
- 4) To measure the association among local MRI signals, network measures, and memory performance across the participants in each of the two subject populations

### **Background and Significance**

Changes in structure and function of the hippocampus, measured by MRI, have been observed in subjects with AD and aMCI. Specifically, AD related hippocampal atrophy has been observed with structural MRI; CBF changes measured by ASL-MRI have been reported in AD; and resting-state fMRI data show that functional connectivity between the hippocampus and other nodes of the default mode network could be modulated by AD. However, the association among the local MRI signals (e.g., the hippocampal CBF), the intrinsic connectivity network integrity (e.g., the functional connectivity between the hippocampus and other default-mode network nodes) and memory performance across the subjects in the aMCI population has not yet been thoroughly studied. We believe that our proposed project, which aims to use a new MRI protocol to quantify the association among hippocampal signals, intrinsic connectivity network integrity and behavioral

measures across aMCI participants, can help characterize the role of the hippocampus for future aMCI research.

### **Year End Progress Summary:**

Our research team has been making good progresses in 1) acquiring MRI data (including ASL-based CBF measures with both quantitative and non-quantitative ASL pulse sequences; T2\*-weighted BOLD fMRI; and structural MRI) from 9 aMCI subjects and 40 non-aMCI subjects (20 with normal cognitive performance and 20 with impaired cognitive performance), 2) establishing a series of post-processing pipelines for analyzing quantitative and non-quantitative CBF data, fMRI data and structural MRI data, 3) developing and evaluating a denoising procedure for reducing noise and improving quality of ASL data, 4) jointly analyzing structural MRI, CBF, fMRI data and behavioral measures, and 5) submitting four NIH R01 grant proposals (in 2019 and 2020) and expecting to secure one of them (in 2020), as described below:

- 1) Acquisition of MRI and behavioral data: Our research team members (Nan-kuei Chen, Ying-hui Chou, Mark Sundman, Yu-Chin Chen) have implemented the proposed protocols in our 3 Tesla human MRI scanner, and acquired MRI and behavioral data from 9 aMCI subjects and 40 non-aMCI subjects (20 with normal cognitive measures; 20 with impaired cognitive performance). Specifically, CBF data were acquired with two types of ASL pulse sequences (Siemens' product non-quantitative PASL sequence and USC-provided quantitative PCASL sequence), resting-state fMRI data were obtained with Siemens' multi-band and parallel EPI sequence, and structural MRI data were acquired with MP-RAGE. Initially we planned to acquire 80 CBF time points to measure dynamic changes of perfusion. However, due to the limitations in both Siemens' and USC-provided ASL pulse sequences, we only acquired 6 CBF time points that are suitable for measuring static CBF map but not dynamic CBF. In order to explore the use of the ASL sequence in measuring dynamic CBF changes, we acquired MRI data both before and after TMS-based neuro-stimulation and then assessed the TMS-induced CBF changes that could be measured with ASL scans.
- 2) Establishment of post-processing pipelines for analyzing CBF and fMRI data: Our research team members (Nan-kuei Chen, Chidi Ugonna) have successfully implemented a series of post-processing pipelines for reconstructing and analyzing the acquired MRI data. Specifically, we have built a Julia-based computer programs that can convert the acquired data to either quantitative CBF map (from USC-provided PCASL protocol) or relative CBF map (from Siemens' product PASL protocol). We have also created Singularity- and Docker-based fMRI analysis pipelines (including quality control; alignment; and other preprocessing tools made available by FSL and Freesurfer etc.), which are installed in both UofA HPC clusters and our recently established XNAT MRI data server. The established pipelines have also been made available to other neuroimaging research groups (including other AAC-funded labs for their ongoing projects).
- 3) Development and evaluation of ASL data denoising procedure: In December 2019, our research team, led by Nan-kuei Chen, has successfully developed and implemented a novel perfusion-matched principal-component-analysis (PM-PCA) algorithm to suppress noise in dynamic ASL scans. Initially our research team planned to evaluate the PM-PCA method in dynamic CBF data acquired from our own 3 Tesla MRI scanner. However, due to the limitations in our existing ASL pulse sequences (that could only acquire a small number of data points), we decided to evaluate the PM-PCA method with dynamic CBF data provided by a collaborator, Dr. Heather Liu. The dynamic CBF data were acquired from the University of Pennsylvania a few years ago, and have been fully de-identified and thus could be shared across research labs. In this evaluation we found that the developed PM-PCA denoising procedure could improve CBF quality and detectability of dynamic perfusion changes.

- 4) Joint analysis of CBF-MRI, fMRI, structural MRI and behavioral measures of aMCI and controls: Our research team (Nan-kuei Chen, Ying-hui Chou, Ashley Stokes, Manoj Saranathan) has just begun to analyze the MRI and behavioral data obtained from our participants. We plan to analyze the data to answer the following research questions. First, do MRI measures (CBF and BOLD) in the hippocampus and other brain regions differ between aMCI subjects and controls? Second, do MRI measures (CBF and BOLD; in the hippocampus and other ROIs) correlate with cognitive performance, even for the non-aMCI population? Using the data processing pipelines we have successfully created; we expect that these two research questions could be answered from our acquired imaging and behavioral data.
- 5) Submission of NIH grant proposals: Our research team members submitted 4 R01 proposals on projects that are highly relevant to this ongoing research topic:
  - a. “Multi-echo and multi-contrast functional MRI using combined spin- and gradient-echo (SAGE) methods” – PI: Ashley Stokes; Sub-award PI: Nan-kuei Chen (submitted in February 2020)
  - b. “High Resolution Anatomical and Structural-Functional Connectivity Imaging of Thalamic Nuclei in Alzheimer's Disease” – PI: Manoj Saranathan; Co-investigators: Nan-kuei Chen and Ying-hui Chou (submitted in October 2019)
  - c. “Accelerated Theta Burst Stimulation for Individuals at a High Risk of Developing Alzheimer's Disease View Text” – PI: Ying-hui Chou; Co-investigator: Nan-kuei Chen (submitted in July 2019)
  - d. “Enhancement of Hippocampal Plasticity Using Repetitive Transcranial Magnetic Stimulation” – PI: Ying-hui Chou; Co-investigator: Nan-kuei Chen (submitted in July 2019). We expect that this Project could be funded starting April 2020.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Transcranial Magnetic Stimulation for Mild Cognitive Impairment.** Ying-hui Chou, ScD, Lee Ryan, PhD, Steven Rapcsak, MD, Nan-kuei Chen, PhD, Phillip Kuo, MD, PhD, Gloria Guzman, MD, Manoj Saranathan, PhD. University of Arizona; Banner University Medical Center; Arizona Alzheimer's Consortium.

### **Project Description:**

Repetitive transcranial magnetic stimulation (rTMS) is a safe and non-invasive neuromodulation technique that is increasingly used in research and clinical practice. Although a number of studies have investigated whether rTMS can improve cognitive functions, a therapeutic rTMS protocol that can be used to attenuate memory loss in people with amnesic mild cognitive impairment (aMCI) is lacking. The purpose of this pilot project is to evaluate the effect of parietal rTMS intervention protocol on memory function in aMCI. The pilot data from our laboratory supports anatomical connection between the hippocampus and the superior parietal lobule (Figure 1). Thus, we will use rTMS to stimulate the superior parietal lobule and examine whether the rTMS can modulate hippocampal function through the transsynaptic connection. Although positive functional connectivity between the hippocampus and the superior parietal lobule has been reported in healthy younger adults<sup>1</sup>, it remains unclear whether the parietal-hippocampal functional connectivity is positive in people with aMCI and how the baseline connectivity moderates the parietal rTMS effect in this population.

### **Specific Aims:**

- 1) To compare the efficacy of excitatory rTMS and inhibitory rTMS vs. sham rTMS on memory function and brain functional connectivity in individuals with aMCI
- 2) To investigate whether baseline brain functional connectivity can be used to predict response to rTMS

### **Background and Significance:**

In our recent meta-analysis that included 13 rTMS studies with a total of 293 individuals with MCI and AD, we reported an overall medium effect size favoring active rTMS over sham rTMS in the improvement of cognitive function (effect size = 0.77,  $p < .0001$ )<sup>2</sup>. Although findings of the meta-analysis are encouraging, the included methodologies are heterogeneous and a specific rTMS protocol is not available. For example, it is unclear which rTMS paradigm (excitatory or inhibitory) has a better therapeutic effect on memory enhancement because inconsistent findings have been reported from both excitatory and inhibitory stimulation paradigms in previous animal models of dementia<sup>3-8</sup>, and in clinical trials for MCI and AD<sup>2</sup>. In this project, we aim to systematically compare the efficacy of excitatory rTMS and inhibitory rTMS vs. sham rTMS on memory function in individuals with aMCI. Moreover, we will assess whether changes in memory function are associated with modulation of memory-related functional networks. Successful achievement of the aims of this proposal would provide direct evidence to support which particular rTMS paradigm (excitatory or inhibitory rTMS) would be more beneficial for memory enhancement in aMCI. Ultimately, this may offer new therapeutic targets that could be applicable not only to aMCI but also to other neurodegenerative diseases.

**Year End Progress Summary:**

We completed the first-year study of 9 individuals with mild cognitive impairment (MCI), with each participants receiving 6 sessions of theta burst stimulation (TBS), yielding a total of 54 TBS sessions within the first year. Our preliminary data analysis indicates that 78% of participants with MCI responded to either excitatory or inhibitory TBS after a single session of stimulation, and the remaining 22% of participants were non-responders. Furthermore, the changes in memory score in response to the TBS were correlated with alterations in hippocampal functional connectivity ( $r = 0.84$ ,  $p = 0.04$ ).

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Development of Small Molecule Dual OX<sub>1</sub>R/OX<sub>2</sub>R Agonist to Explore the Role of Orexin System in Alzheimer's Pathology.** Kevin Gaffney, PhD, Kathy Rodgers, PhD. University of Arizona; Arizona Alzheimer's Consortium.

### **Specific Aim:**

In *Specific Aim 1*, we will generate a number of analogs and molecular hybrids between Yan 7874, Dag 26, & suvorexant and test both their agonist activity and affinity on both orexin receptors, the OX<sub>1</sub>R and OX<sub>2</sub>R, to explore the structure activity relationship (SAR) of small molecule orexin agonists. We will then use those insights to develop a potent, non-cytotoxic dual OX<sub>1</sub>R/OX<sub>2</sub>R agonist for future assessment in Alzheimer's disease (AD) pre-clinical models.

### **Background and Significance:**

Alzheimer's disease (AD) is a devastating, progressive neurodegenerative disease that affects over 5.5 million Americans. AD is the 6<sup>th</sup> leading cause of death in the US and is the most common cause of dementia making it one of the greatest therapeutic needs of the 21<sup>st</sup> century. Unfortunately, despite tremendous efforts, the development of new therapeutics for AD suffer from the highest failure rates of any disease state. As a result, the identification of novel therapeutic targets for AD are of the utmost importance.

One of the symptomatic hallmarks of AD is sleep disturbances. Sleep impairment is increasingly seen as a marker of the prodromal phase of AD, mild-cognitive impairment. During sleep, there is an increase in cerebrospinal fluid flow which removes toxic metabolites and waste from the brain. Further, sleep deprivation has been shown to increase CNS levels of amyloid  $\beta$  (A $\beta$ ) and tau. Sleep is modulated in part by the activation of the orexin receptors OX<sub>1</sub>R and OX<sub>2</sub>R by their ligands orexins A and B. In the Tg2576 mouse model of AD, treatment with dual OX<sub>1</sub>R/OX<sub>2</sub>R antagonist almorexant decreased A $\beta$  levels and A $\beta$  plaque formation. However, we hypothesize that orexin receptor agonists would decrease pathology in AD as orexin peptides have been shown to be neuroprotective in models of AD, age-related cognitive decline, and Parkinson's. In order to test this hypothesis, in this proposal we seek to: (i) better understand the small molecule SAR for both the OX<sub>1</sub>R and OX<sub>2</sub>R; (ii) use these insights to develop a potent, non-cytotoxic dual OX<sub>1</sub>R/OX<sub>2</sub>R agonist. The successful completion of these goals will allow us to assess the efficacy of OX<sub>1</sub>R/OX<sub>2</sub>R agonists in AD pre-clinical models.

### **Year End Progress Summary:**

In execution of our ultimate goal of developing potent, non-cytotoxic a dual OX<sub>1</sub>R/OX<sub>2</sub>R agonist, we have carried out a number of computational studies to model the interactions of existing OX<sub>1</sub>R and/or OX<sub>2</sub>R agonists on their respective receptors. Based on these computational insights, we have designed a focused array of potential OX<sub>1</sub>R/OX<sub>2</sub>R agonists. We are currently nearing the completion of the synthesis of this focused OXR array and will carry out testing of this series of molecules in the next few months.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Improving clinical neuropsychological assessment of subtle cognitive decline and mild cognitive impairment.** Matthew D. Grilli, PhD, Jessica R. Andrews-Hanna, PhD, Matthias Mehl, PhD, Lee Ryan, PhD, Matthew Huentelman, PhD. University of Arizona; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

This project is designed to improve our assessment of cognitive and functional alterations associated with prodromal Alzheimer's disease (AD). To our knowledge, it will be the first to use naturalistic assessments to quantify real-world thinking patterns in individuals with mild cognitive impairment (MCI) or subtle cognitive decline (SCD).

### **Specific Aims:**

- 1) To test the hypothesis that specificity of everyday thought and social conversation is compromised in individuals with MCI and individuals with SCD. We predict that relative to cognitively healthy older adults, individuals with MCI or SCD will exhibit autobiographical thoughts that are less spatiotemporally specific and more present-bound.
- 2) To uncover the extent to which alterations in everyday thought and social conversation are related to objective and subjective evaluation of prospective memory, in both cognitively healthy older adults and individuals with MCI or SCD. Specific and future-oriented autobiographical thoughts are important for prospective memory, which is our ability to remember to follow through with delayed goals. Many everyday tasks require "remembering to remember" and thus it is not surprising that impaired prospective memory has been linked to reduced functional status in individuals with MCI. We predict that less spatiotemporal specific and less future-oriented thought in everyday life will be associated with reduced prospective memory among older adults, including individuals with MCI or SCD.
- 3) To develop a pipeline for longitudinal assessment of older adults, including individuals with MCI or SCD, using a combination of naturalistic and standard neuropsychological tools.

### **Background and Significance:**

The ability to turn our thoughts inward is fundamental to remembering the past, contemplating the future, and engaging socially. Only recently have we begun to realize that autobiographical thought can be compromised in individuals at increased risk for AD and related dementias, including in individuals with MCI. However, our understanding of how MCI affects autobiographical thought is based largely on lab-based tasks that poorly represent the complexities of everyday life, with no studies of MCI objectively evaluating autobiographical thought in the real world. Mobile smartphone tools, which can sample real-life thoughts and cognition in action, can help close this gap in knowledge and provide novel insights into the real-world impact of cognitive impairment, such as how cognitive difficulties affect prospective memory, a key determinant of functional independence. Naturalistic assessment tools also have the potential to improve clinical neuropsychological assessment by not only being cost-efficient and convenient tools, but also by improving diagnostic accuracy, enhancing both sensitivity and specificity to abnormal cognition by uniquely tapping the impact of everyday environments over time. This proposal is innovative because it will be the first to measure the extent to which MCI and SCD are associated with alterations in autobiographical thought and language in the real

world using novel naturalistic assessment tools developed by our collaborative research team. It also extends our theoretical model of AD risk by evaluating previously untested prodromal factors. Moreover, this project will be the first to evaluate whether naturalistic assessment tools provide insight into daily thought patterns that contribute to prospective memory performance, which is commonly impaired by MCI.

**ACHIEVE.** This project builds on “Aging, Cognition, and Health: An Interdisciplinary, Ecologically-Valid Experiment” or ACHIEVE, which is a collaborative project on cognitive aging that brings together multiple labs in the Psychology Department at UA. This project includes a Neuropsychology Core (PI Grilli), Naturalistic Assessment Core (PI Andrews-Hanna), and Neuroimaging Core (PI Ryan). The Neuropsychology Core is responsible for screening and cognitive testing for all AAC Projects in ACHIEVE.

### **Year End Progress Summary:**

- 1) We remain highly active in recruitment and enrollment in ACHIEVE. In the past year, the Neuropsychology Core has screened 115 individuals. We have completed neuropsychological testing and genotyping for 88 of these individuals. These individuals have been directed to ACHIEVE projects based on study eligibility, with ongoing enrollment.
- 2) Both smartphone apps have been fully developed and updated, piloting for all in-lab and naturalistic methods used in the present project is complete, and data collection is ongoing.
- 3) As MPIs, Grilli and Andrews-Hanna, along with Co-I’s Mehl and Huentelman, submitted an R01 to NIH/NIA that builds on the aims of this project and would further investigate the utility of naturalistic assessment of cognition and Alzheimer’s disease risk in older adults.
- 4) Consistent with Aim 3, we created a pipeline for longitudinal assessment, which involves tracking participant interest in remaining engaged and contacted for future studies. Participants are scheduled to repeat neuropsychological testing every 1.5-2 years.
- 5) In accordance with the overarching goal of this project, Drs. Grilli, Andrews-Hanna, and Mehl contributed to a study examining the naturalistic assessment of autobiographical memory and future thinking in older adults (Wank et al., under review), which was invited to be part of a special issue in *Frontiers in Human Neuroscience*.
- 6) Dr. Andrews-Hanna and Dr. Grilli have been invited to submit a piece to *Current Directions in Psychological Science* that includes a discussion of naturalistic assessment of cognition (first draft deadline 4/15/2020).
- 7) For Dr. Grilli’s lab, participants screened and tested through the Neuropsychology Core supported two dissertation projects, two poster presentations at the International Neuropsychological Society Annual Meeting (February 2020), and two poster presentations at the Cognitive Aging Conference (April 2020).

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Defining the effect of a rare mutation S305Y on CD44 causing Alzheimer's disease.** May Khanna, PhD, Matt Huentelman PhD. University of Arizona; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

The goal of the project is to begin to dissect the effect of a rare mutation (S305Y) in CD44 that has been associated with a patient with young onset Alzheimer's disease

### **Specific Aims:**

- 1) To define the interaction between CD44 (wt and S305Y) with proteins using peptides pull down followed by mass spectrometry
- 2) To express CD44 in the presence and absence of mutation and obtain preliminary data for CRYO-EM structure elucidation

### **Background and Significance:**

CD44 is a transmembrane glycoprotein, also known as P-glycoprotein 1. CD44 is a multifunctional cell surface adhesion receptor. The interaction with appropriate extracellular matrix ligands promotes migration of cells. CD44 was additionally identified as a stem cell marker. CD44 is encoded by a single gene and is ubiquitously expressed. The expressed protein has a molecular weight that ranges from 85-200 kDa depending on the splicing pattern. There is one form known as CD44s which has a molecular weight of 85-90 kDa, and the focus of our study. There are also splicing patterns that yield protein with varying molecular weight that have been found upregulated in Alzheimer's disease (Pinner *et. al.*).

CD44 is an inflammation-related gene with multiple splice variants expressed at the cell surface. The splice variants were investigated in post-mortem brains of Alzheimer's patients. It was determined that the following were detectable: CD44S (does not contain any alternative exon) and CD44V3, CD44V6 and CD44V10 splice variants. All these forms were significantly higher with patients with Alzheimer's compared to normal patients. Immunohistochemistry revealed that CD44S was localized in neuritic plaques and astrocytes whereas CD44V3, CD44V6 and CD44V10 were mostly neuronal. CD44V10 and CD44V6 were induced by A $\beta$  peptide in neuroblastoma and primary neurons. It was further shown that targeting CD44V10 with an antibody protected cells from A $\beta$ -induced toxicity.

Recently, the laboratory of Dr. Matt Huentelman at TGEN in Phoenix discovered a rare mutation associated with young onset Alzheimer's disease (YOAD). The mutation is located in the extracellular portion of CD44 at position 305. This mutation is not part of known splice variants associated with Alzheimer's. The protein that contains this mutation is upregulated in several parts of the brain. The focus of this grant is on this mutation and the effect on CD44.

### **Year End Progress Summary:**

- 1) We synthesized two peptides that express the region that has the mutation at position 305 and that were wild-type CD44. These peptides are short peptides with a tag that can be used for pull-down.

- 2) We attempted the pull-down with cell cultures at first in order to optimize the conditions, but were not able to get enough proteins from the pull down experiments. We then optimized the pull-down of CD44 peptide and CD44 peptide with mutations using rat brain samples that were then subjected to mass spectrometry. We are currently awaiting results from the mass spec to compare pull down from the wild-type and mutant peptide. Based on the silver staining of the peptide pull-downs, we are confident that there are differences between the two peptides. This will be critical to determine if there are any alterations of PPI between the disease causing mutation and wild-type CD44.
- 3) We have begun expressing CD44 with a tag in order to purify this protein. This has been quite challenging. We have tried a Histidine tag (our traditional purification methods), but this seems to cause issues with expression with CD44. We are currently trying an HA tag version of CD44 and are trying the purification in the presence

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Mechanisms of Neuroprotection by Mas Agonists.** Kathleen Rodgers, PhD, Fei Yin, PhD, Ted Trouard, PhD, Ashley Stokes, PhD. University of Arizona; Barrow Neurological Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

We *hypothesize/propose* that the anti-inflammatory and redox modifying effects of our small molecule Mas agonist (RASRx1902) will have a beneficial effect in reducing the risk and delay in the progression of Alzheimer's Disease (AD); and that its regenerative properties will assist in repairing brain damage due to AD. No human studies have been conducted using Mas agonists to treat AD, making it an attractive novel therapeutic target.

### **Specific Aims:**

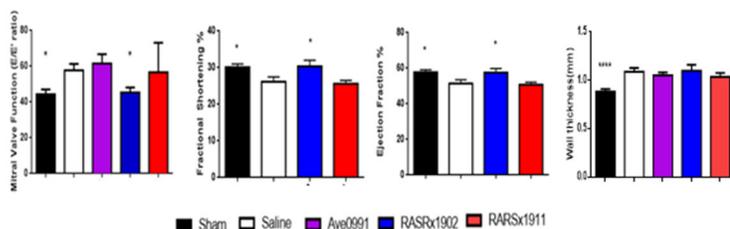
- 1) To evaluate the mechanisms by which RASRx1902 improved cognitive function and reduced oxidative stress in vivo. These studies will focus on mitochondrial function and endothelial dysfunction.
- 2) To dissect direct effects of RASRx1902 on microglial function after exposure to toxic stimuli in vitro.
- 3) To conduct pilot studies on blood brain barrier integrity and cerebral blood flow in animals that underwent transaortic constriction surgery.

### **Background and Significance:**

Persistent activation of the A-II/AT<sub>1</sub>R axis results in extensive cerebrovascular remodeling, inflammation, and oxidative stress (OS) leading to neurovascular uncoupling and disruption of the blood brain barrier (BBB)<sup>1,2</sup>. The protective arm of RAS, comprised of the Mas receptor and its ligand A(1-7), potential to treat AD. In AD patients, serum and brain ACE2—the main A(1-7)-producing enzyme—levels are reduced compared to control subjects<sup>3</sup>. In these brain, ACE2 was inversely correlated to A $\beta$  levels and phosphorylated tau pathology<sup>4</sup>. These trends were mirrored in animal mouse models of AD. In senescence-associated mouse prone 8 (SAMP8) mouse model of sporadic AD, brain levels of A(1-7) were found to be low while brain tau hyperphosphorylation levels were elevated, a trend also seen in P301S mice, a model of pure tauopathy<sup>5</sup>. The effects of Ang-(1-7) treatment on AD was tested in the 5xFAD mouse which develops amyloid deposition and cognitive deficits at as early as 2 months of age<sup>6</sup>. Intracerebroventricular (ICV) infusion of A(1-7) ameliorated cognitive impairment and increased cerebral blood flow reactivity in these mice. ICV treatment of stroke prone spontaneously hypertensive rats with A(1-7) increased neuronal survival, neurological status, neuronal survival, and overall survival while decreasing the incidence of hemorrhages, indicating a reversal of micro-vessel dysfunction<sup>7</sup>.

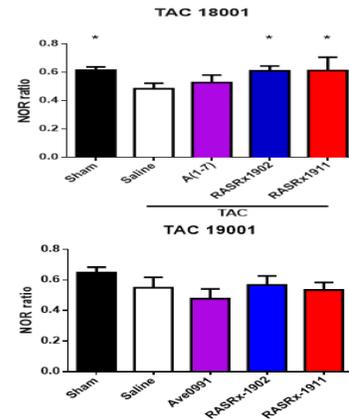
### **Year End Progress Summary:**

1a) TAC impairment of Cardiac Function. In 8-10wk male C57Bl/6J, TAC surgery induced marked dysfunction in standard cardiac measures of left ventricular function, which were rescued by 1902 treatment in each case. As a within study control, ventricular wall thickness was measured to ensure that treatment groups were not

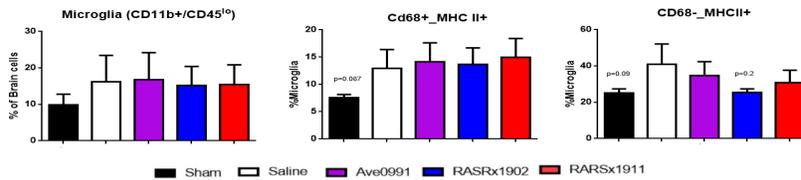


in end-stage heart failure, indicated by wall thinning, and showed a consistent increase among all surgical groups (Figure 1).

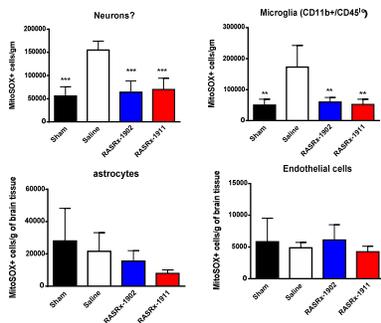
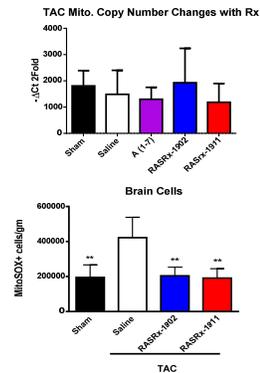
1b) Cognitive Impairment and Rx. Effect Decline in cognitive function was assessed 10wk-post surgery via novel object recognition (NOR). Controlling for anxiety and decreased activity, analysis of our data (TAC19001) showed a trend that is comparable to data generated in the preliminary iteration of this study (TAC18001) (Figure 2). In the current study, loss of significance is thought to be due to a difference in manual vs automated scoring and a decrease in the overall TAC gradient of the surgical groups, that produced a less severe constriction and thus failed to produce the cognitive impairment previously demonstrated (Data not shown).



1c) Microglial Function in Disease Pathology. Given the critical role of microglia in maintaining brain health, and the growing body of evidence showing its destructive capacity in chronic states, it was hypothesized that microglia are the primary driver of pathology resulting in the cognitive deficits seen. Analysis showed that although overall numbers were not increased, TAC induced a strong increase in antigen presenting microglia, that was rescued with 1902 treatment, when not expressing CD68 (Figure 3).



1d) Mechanisms of RASRx-1902 treatment effect. Composite RNA isolated from hippocampal sections were outsourced for RNAseq analysis. Results were examined with Ingenuity Pathway Analysis and significant changes from surgery were seen in a downregulation of mitochondrial oxidative phosphorylation and an upregulation in apoptosis and oxidative stress, which were reversed with 1902 treatment (Data not shown). Investigation of the mechanisms behind this therapeutic effect was revealed to be a maintenance of existing mitochondrial function rather than an induction of mitochondrial biogenesis (Figure 4).

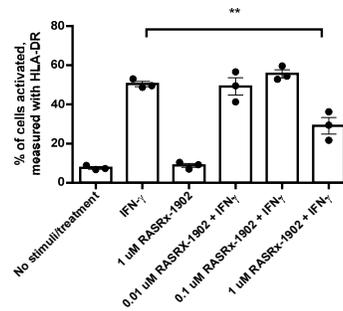
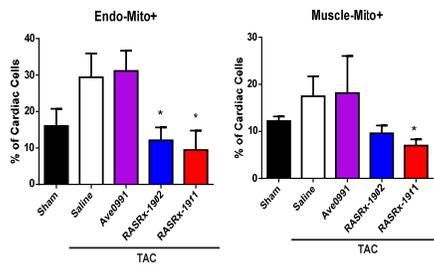
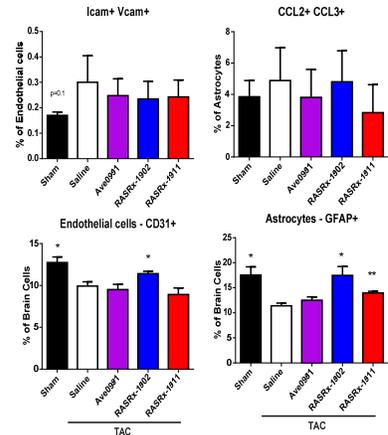


1e) Cell Specific Changes with TAC and RASRx-1902 Rx. Given the general increase of mitochondrial dysfunction seen in the brain, further investigation into the specific cell types affected was warranted. A screen of all known major CNS cell types revealed TAC-induced mitochondrial dysfunction to be sequestered to microglia and possibly neurons. Surprisingly, endothelial cells and astrocytes did not show such dysfunction (Figure 5).

1f) Glial and Astrocyte viability and phenotypic changes with TAC. Given that TAC is a cardiac model, we were prompted to further investigate the exclusion of astrocytes and endothelial cells showing mitochondrial dysfunction. Studies indicate that areas of amyloidosis are localized to where endothelial cells express ICAM and VCAM. Similarly, expression of the chemo-attractants

MCP-1 and MIP-1a, possibly on astrocytes, can contribute to an increase in inflammation and progression of disease pathology. Testing of this hypothesis demonstrated that although TAC failed to induce the phenotype changes seen the literature, it did affect the viability of these cell types, showing significant reduction in the total percentage, that was rescued with treatment (Figure 6).

1g) Oxidative stress may play a role in TAC-induced cardiac dysfunction. Throughout these experiments, we are demonstrating that mitochondrial dysfunction plays a major role in TAC-induced AD-like pathology. In this vein, we decided to evaluate the heart itself for the same effects. Compared to the surgical group (Saline), data showed a significant decrease in dysfunction with treatment for both cardiac endothelial cells and cardiomyocytes (Figure 7 below).



## 2) RASRx-1902 Produces Critical Dose reduction of Microglial Activation under Stress Stimuli in vitro.

To further demonstrate the anti-inflammatory modifying effects of 1902 treatment, the immortalized microglia cell line, HMC3, was exposed to various activation stimuli and treated with a range of RASRx-1902 concentrations. With HLA-DR serving as a marker for microglial activation, HMC3 cells showed significant and consistent activation upon 48hr IFN-gamma exposure, to be rescued by 1902 treatment at 1  $\mu$ M (Figure 8 above).

## 3) GD-enhanced MRI to evaluate blood brain barrier integrity and cerebral blood flow.

A preliminary study using three (3) Sham and three (3) TAC mice is currently underway using t1 star weighted dynamic susceptibility contrast (DSC) to indicate watch in and wash out of the contrast dye through vasculature, thus acting as a measure for blood volume and flow. A t2 star weighted dynamic contrast enhanced (DCE) will also be used over a longer period, as a measure of total accumulation of the contrast dye and thus, a measure of BBB integrity.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Contextual retrieval impairment in self-defining autobiographical memories as an early indicator of risk for AD.** Lee Ryan, PhD, Matthew D. Grilli, PhD, Jessica Andrews-Hanna, PhD, Matthias Mehl, PhD, Matthew Huentelman, PhD. University of Arizona; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

The proposed project is designed to improve our assessment of cognitive and functional alterations associated with preclinical Alzheimer's disease (AD) by identifying age-related memory changes that are linked to risk factors for AD including APOE e4 status, family history of AD, or subtle cognitive decline (SCD). Our recent study suggests that at-risk and non-risk older adults differ in the number and quality of perceptual and spatial-temporal details that are retrieved during autobiographical memory<sup>1</sup>. These differences may be due to alterations in medial temporal lobe (MTL) circuits mediating detailed versus holistic memory retrieval<sup>2</sup>. Importantly we hypothesize that circuit alterations may be particularly evident for some forms of autobiographical memory. This project builds upon the ACHIEVE project, which links several laboratories working collaboratively to build ecologically-valid, interdisciplinary evaluations of aging and AD risk.

### **Specific Aims:**

1. To test the hypothesis that individuals at high risk for AD, including SCD, retrieve fewer perceptual and spatial-temporal details for specific events relative to low-risk older adults. We expect that this difference may be particularly evident during retrieval of a special class of autobiographical memories referred to as "self-defining" memories – event memories that are typically well-rehearsed and imbued with significant personal meaning.
2. To determine whether the number and quality of retrieved event details are related to performance on perceptual and memory tasks that known to be mediated by direct and indirect MTL memory circuits. We predict that retrieval of spatial-temporal detail will be related to context-specific object memory (pattern completion) but not object recognition (pattern separation).
3. To determine whether fMRI activation during pattern completion and pattern separation tasks will be associated with risk factors for AD.

### **Background and Significance:**

A recent study by our group<sup>1</sup> demonstrated that cognitively healthy older adults who carry the APOE e4 allele provide fewer perceptual and spatial-temporal details when describing autobiographical events, despite performing similar or even better than non-carriers on conventional neuropsychological tests of memory. This finding suggests that autobiographical event retrieval may provide a particularly sensitive and specific marker of neural changes associated with preclinical AD. In a recent theoretical paper<sup>2</sup>, we suggested that there are two parallel but relatively independent memory retrieval pathways, both relying upon contributions from perirhinal cortex and parahippocampal cortex. The "coarse" pathway creates a global representation of the environment that includes gist-like information regarding the spatial-temporal relations among various disparate components of a memory. The second pathway, the "detail" pathway, provides information on the combinations of specific features of environment, and is utilized whenever there is ambiguity or unfamiliarity in the environment. In that case, the detail pathway takes precedence and provides additional feature information that disambiguates experience. Based on data from animal models and human studies, we hypothesized that the

detail pathway is impaired in normal aging due to the deposition of tau pathology, while the coarse pathway remains intact. In contrast, individuals with preclinical Alzheimer's pathology results in increases in both tau and a beta deposition in regions that will damage the coarse pathway. Damage to the coarse pathway will likely result in a loss of easy access to the disparate components of an episode, since retrieval of a single component of the memory will no longer lead to relatively automatic reinstatement of the rest of the memory.

Our laboratory recently published data demonstrating that older adults are impaired relative to young adults on visual "pattern separation" tasks that are hypothesized to rely on the detail pathway. Additionally, we showed that older adults utilize context to the same degree as older adults in order to support object recognition and associative memory<sup>3</sup>, suggesting that the coarse pathway remains relatively intact with age. However, new data suggest that context-specific recognition may be impaired in older adults with genetic and cognitive risk for AD (Ryan et al., in preparation). At-risk older adults are less likely than low-risk individuals to rely on context to recognize previously studied objects, and they make fewer false recognition errors when a context is repeated. We hypothesize that impairment in context-specific recognition will be related to a decrease in the spatial-temporal contextual details retrieved from event memories, since both these tasks may be mediated by the MTL coarse pathway.

The difference in contextually-dependent retrieval associated with AD risk may be particularly apparent for some forms of event memories. While autobiographical events from our past that are considered personally relevant tend to be recalled in detail, memories for events that are considered ***integral to one's personal identity*** are especially vivid and rich in perceptual and spatial-temporal detail. These "self-defining memories" (SDMs) are often shared over many years with family and friends, and they are subjected to intense personal introspection. SDMs are characterized by a consistent narrative built around the event that incorporates not only the event-specific details of the event, but also the meaning derived from the event and connections to other relevant life experiences. Compared to young adults, older adults, on average, tend to provide more meaning-related context while recalling SDMs<sup>4</sup>, and SDMs may be embellished with even richer semantic/contextual information. However, Martinelli et al.<sup>5</sup> reported that older adults with mild cognitive impairment recalled fewer SDMs, and those memories were particularly lacking in detail, suggesting that the resilience of SMDs to normal aging may not persist in the presence of pathology associated with early AD.

The goal of the proposed project is to further our understanding of how aging and risk for AD differentially impacts autobiographical retrieval. We intend to examine whether cognitively healthy older adults with increased risk for AD have greater difficulty accessing SDMs, and describe less meaning-related content and spatial-temporal context for these memories. We predict that this ability will be directly related to utilization of context during laboratory-based context-dependent recognition tasks that rely on pattern completion, and will be reflected in changes in the patterns of fMRI activation in medial temporal lobe regions during these tasks.

#### **Year End Progress Summary:**

Data collection for this project is well underway. To date we have collected behavioral data from nearly 60 older adults and young adults who have been characterized for APOE e4 status. These behavioral studies have yielded interesting results suggesting that older e4 carriers and non-carriers utilize detail in memories to differing degrees, and have differential patterns of errors as a result. These data are accepted for presentation at the upcoming Conference on Aging in

Atlanta Georgia in April, and a manuscript is being prepared for publication. Additional data from the study supported two recent publications, listed below:

Lawrence, A., Cardoza, J., Ryan, L., (in press). Medial temporal lobe regions mediate complex visual discriminations for both objects and scenes: A process-based view. *Hippocampus*.

Memel, M., Wank, A.A., Ryan, L., Grilli, M.D. (2019) The relationship between episodic detail generation and anterotemporal, posteromedial, and hippocampal white matter tracts. *Cortex*, 123:124-140.

The neuroimaging portion of the project is ongoing. A novel fMRI task that targets context-dependent retrieval of episodic memories has been developed and piloted extensively on both young and older adults. Scanning is underway and we anticipate that data collection will be completed by the end of June. Two of the researchers are already working on an R01 to be submitted in fall, 2020 based on these experiments.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Establishing pipelines data sharing and image analysis for cognitively healthy older adults at the University of Arizona.** Lee Ryan, PhD, Nan-Kuei Chen, PhD, David Labiner, MD, Gene Alexander, PhD, Carol Barnes, PhD, Ted Trouard, PhD, Roberta Brinton, PhD, Matt Huentelman, PhD, Thomas Beach PhD. University of Arizona; Translational Genomics Research Institute, Banner Sun Health Research Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

This project is designed to establish large-scale data sharing and image analysis pipelines at the University of Arizona that will greatly enhance our ability to build large-scale datasets across laboratories and share data with the larger research community. In addition, we are establishing standardized procedures for biomarker collection (CSF and blood) from cognitively healthy older adults. This effort will extend existing databases available from cognitive impaired older adults by providing biomarkers from well-characterized older adult control participants who are available for future studies.

### **Specific Aims:**

- 1) To collect and bank biomarker specimens using standardized procedures, including blood and CSF, from well-characterized cognitively healthy older adults.
- 2) To build a database for sharing data from cohorts of cognitively healthy older adults across multiple laboratories within the Arizona Alzheimer's Consortium, including neuropsychological testing, neuroimaging, and biomarker data.
- 3) To establish a pipeline for standardized state-of-the-art MRI analysis to optimize and streamline MRI analysis methods.

### **Background and Significance:**

Understanding the variability of cognitive trajectories in normal and pathological aging requires data from large numbers of participants who are well characterized. Many of the most promising approaches to early detection and predicting cognitive decline in otherwise healthy older adults involves biomarkers from blood, CSF, and neuroimaging, among others. The complexity and high cost of collecting large-scale datasets with these types of measurements highlights the importance of sharing data across laboratories. The funds for this project will be used to collect biomarkers including blood, CSF, and neuroimaging data from cognitively well-characterized older adults without a diagnosis of dementia, using standardized protocols that were established in 2018-2019. Drawing on expertise at UA and our partner institutions within the AAC, we will expand our database for sharing standardized measurements that will be made available to all AAC researchers. Additionally, we will establish a pipeline for standardized state-of-the-art MRI analysis to optimize and streamline MRI analysis methods.

### **Year End Progress Summary:**

We have made considerable progress on all Aims in the project. Computing facilities have been built adjacent to the research MRI which are available to all AAC research. Libraries of images from two laboratories have been catalogued and stored as test cases for data sharing, using the XNAT central repository. XNAT is a publicly available imaging data repository, funded by NIH. This will allow us to share imaging libraries with researchers not only within the AAC but across the US, and will greatly enhance our ability to engage in large-scale studies of aging.

Additionally, MRI analysis methods for volumetry and tractography have been established using the HPC cluster. A two-day workshop is planning for April 27-28<sup>th</sup> where researchers will learn how to access the pipeline, utilizing the high performance computing facilities, for data analysis, and how to store and share data on Cyverse. In addition to funding from AAC, this project has obtained additional funding from the University of Arizona's President's Office in conjunction with the University of Arizona 2018 Strategic Plan.

Regarding biomedical samples, we have a full protocol in place for collecting and processing blood samples for long-term storage, with wet lab space and freezers available adjacent to the MRI scanner. We expect to bank samples from 60 older adults by June 2020. For CSF draws, we are now consulting with Dr. Ali Atri at Banner Sun Health Research Institute on a protocol for collecting and banking CSF. Dr. Atri currently has a grant to develop materials and methods for increasing success rates in obtaining CSF samples from volunteers.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**MRI and automated segmentation of thalamic nuclei for Alzheimer's Disease (AD).** Manojkumar Saranathan, PhD, Gloria J. Guzman, MD, MSc, Nan-kueh Chen, PhD, Ying-Hui Chou, PhD, Geoffrey Ahern, MD, PhD, Alex Hishaw, MD, Eric Reiman MD. University of Arizona; Banner Alzheimer's Institute; Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) To perform volumetry of thalamic nuclei and hippocampal subfields on a cohort of normal controls and patients with mild cognitive impairment (MCI).
- 2) To validate the performance of the thalamic nuclei segmentation on conventional MPRAGE and perform a retrospective analysis on an age-matched cohort of normal controls, MCI, and AD patients taken from the ADNI database.

### **Background and Significance:**

Most MRI studies of AD document volumetric changes of whole hippocampi. The role of the thalamus in AD has been ignored despite reports that thalamic structures like the antero-dorsal nucleus are affected<sup>1</sup>, due to poor visualization of thalamic nuclei<sup>2</sup>. There is increasing evidence<sup>6</sup> that AD might affect deep brain structures in a more targeted fashion (e.g. CA1). Investigating volume changes in thalamic nuclei and hippocampal subfields might be more sensitive to the disease process, provide a more detailed picture of disease progression, and improve sensitivity/specificity compared to whole volume analyses.

We have developed new methods for automated segmentation of thalamic nuclei for 7T and, recently, for 3T MRI (AAC funding from 2018-2019) and optimized for 7T and 3T MRI<sup>3</sup>, a white-matter nulled (WMN) MPRAGE<sup>4</sup> sequence which provides excellent delineation of the thalamus boundary and improved intra-nuclear contrast. This sequence is also motion robust, ideal for imaging patients with dementia. We have developed THOMAS, a fast segmentation method for delineation of thalamic nuclei<sup>5</sup> in 10 minutes vs. manual processing time of several hours.

Lastly, a large database of MRI from clinically confirmed MCI & AD and healthy control subjects are available in the ADNI database. However, they use conventional MPRAGE and not the WMN MPRAGE that THOMAS requires for thalamic segmentation. We are developing a new version of THOMAS that can segment conventional MPRAGE, giving us access to vast amounts of existing patient data. This can be used to assess if thalamic nuclei volumes are selectively impacted in MCI and AD and follow it across time.

### **Year End Progress Summary:**

**Aim 1.** Healthy aging control and MCI subjects were acquired on the 3T Siemens MRI research scanner. These were augmented with clinical data obtained on 3T Siemens MRI clinical scanners from patients with clinically diagnosed MCI and AD status. THOMAS was used to analyze the thalamic nuclei from the WMN MPRAGE sequence, which was added to the protocols. A total of 20 AD patients, 16 MCI patients, and 13 healthy controls were analyzed. For AD patients, in addition to pulvinar and centromedian, ventral nuclei such as ventral anterior, ventral lateral anterior and ventral posterior lateral were significantly atrophied compared to healthy controls after adjusting for age and intra cranial volumes (ICV). For MCI patients, only ventral lateral

anterior showed atrophy compared to healthy controls. Both AD and MCI showed whole thalamus atrophy as expected. These results, especially the lack of change in anteroventral and mediodorsal nuclei (limbic nuclei involved in episodic memory) should be interpreted in the light of the small sample size. We continue to acquire more data to a final goal of about 30 subjects per cohort to achieve statistical power for all nuclei. Hippocampal subfield analysis is also currently in progress to determine if the changes in thalamic volume are correlated with changes in hippocampal subfields. *Note that these results are the first in literature to document changes in specific thalamic nuclear volumes in MCI and AD (as opposed to whole thalamus which has been documented in literature)*

**Aim 2.** To increase the usability, we adapted THOMAS to process conventional MPRAGE (as opposed to WMN MPRAGE which is a special sequence and not available at all MRI scanners). We utilized data available from 7T (Stanford collaboration with Dr. Brian Rutt) where both MPRAGE sequences were acquired on patients with multiple sclerosis and healthy controls. These data were also manually segmented by a neuroradiologist using the Morel atlas as a guide to enable direct comparison of the proposed modified method to THOMAS on Wmn MPRAGE. Dice indices of 0.8 or higher were achieved for larger nuclei (pulvinar, mediodorsal) and 0.7 or higher for most remaining nuclei (LGN, MGN, ventral anterior) with anteroventral nuclei achieving a Dice of 0.62.

This initial validation gave us the confidence to perform a larger retrospective analysis of the ADNI database. First, we attempted to analyze 1.5T data which was of poor quality (motion, poor SNR) and variable (inconsistent protocols across sites). We then switched to 3T data, which was of higher quality and much more consistent. A cohort of about 150 patients equally divided into healthy controls, AD, and MCI subjects was downloaded and analyzed using custom developed software. After correction for intracranial volumes (ICV) and age, we found statistically significant (FDR corrected p-values) reductions in whole thalami as well specific nuclei which different across hemispheres (left vs. right) and with disease (MCI vs. AD). For AD, only pulvinar was smaller in the left side compared to healthy controls while pulvinar, centromedian, MGN and mediodorsal nuclei were smaller in the right side compared to healthy controls. For MCI, pulvinar and centromedian were smaller in the left side, while MGN and centromedian were smaller in the right side, compared to healthy controls. We are currently trying to correlate neuropsych scores also available on these patients to thalamic nuclear volumes to see if there are any correlations. We are also trying to reconcile discrepancies in the results between this cohort using conventional MPRAGE and our earlier smaller cohort using WMN MPRAGE.

An R01 proposal (10028636 ID, total direct \$360,578.00) titled “High Resolution Anatomical and Structural-Functional Connectivity Imaging of Thalamic Nuclei in Alzheimer's Disease” with preliminary data from the above analyses was submitted to NIA/NIBIB in October, which is under review. A manuscript is under preparation with the thalamic nuclei volume analysis from Aims 1-2 to be submitted to Neuroimage Clinical end of March.

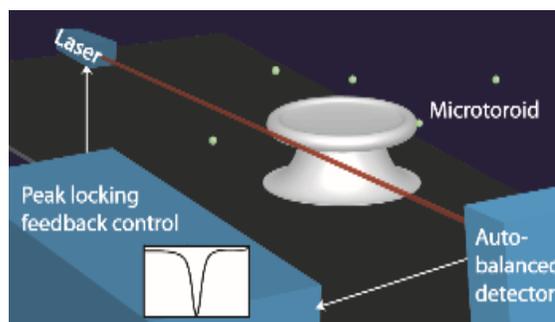
# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Determining levels of Alzheimer's biomarkers in postmortem cerebral spinal fluid and serum samples.** Judith Su, PhD, Gene Alexander, PhD, Thomas Beach, MD, PhD. University of Arizona; Banner Sun Health Research Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

We have developed a technique known as FLOWER (frequency locked optical whispering evanescent resonator) (Figure 2) that can detect low concentrations of molecules down to the single molecule limit without requiring the use of labels such as fluorescent or radioactive tags.<sup>1</sup> We are currently evaluating the ability of FLOWER to test for the Alzheimer's disease (AD) biomarkers amyloid beta and tau in both cerebrospinal fluid (CSF) and serum/plasma. There are potential benefits for applying FLOWER to both types of samples. For CSF, FLOWER offers greater sensitivity that could be more reliable and robust, cheaper, and easier to reproduce across labs. Furthermore, because of its particularly high sensitivity, FLOWER offers the potential to detect Alzheimer's disease biomarkers in serum and/or plasma, which can be more easily collected from participants with lower risk than the collection of CSF. This is especially conducive for repeated measurements. The serum/plasma detection can be directly assessed against the CSF markers to help validate the measures with established markers that reflect deposition in brain. The objective of this one-year pilot project is to obtain the necessary preliminary data that demonstrates the feasibility of our approach and will form the basis for an R01 proposal, as well as for publications to further support this research plan. In previous pilot project, we focused on detecting the AD proteins, amyloid beta and tau using FLOWER. This is related to an R03 (PI: Su; R03AG055020) we were awarded. In the current pilot project proposal, we will focus on benchmarking our amyloid beta detection results in CSF and serum using ELISA.



*Figure 2. FLOWER schematic. In the FLOWER detection scheme, microtoroid optical resonators are used in conjunction with frequency-locking feedback control and balanced detection to improve the signal-to-noise ratio of the system. As a result, single-macromolecule detection becomes possible.*

### **Specific Aims:**

- 1) Conduct a hemoglobin assay to screen out hemoglobin contaminated patient samples.
- 2) Perform an ELISA assay for amyloid beta on 100 CSF postmortem patient samples from patients with varying degrees of AD severity.
- 3) Perform an ELISA assay for amyloid beta on 100 serum postmortem patient samples from patients with varying degrees of AD severity.

### **Background and Significance:**

FLOWER has achieved a signal to noise ratio of 5 using an anti-IL-2 antibody layer immobilized on a microtoroid to specifically capture IL-2. Direct detection of biomarkers such as amyloid beta is possible because the binding of proteins to antibodies on the surface of the microtoroid produces a detectable optical thickness change. Demonstrating the feasibility of our concept for ultra-sensitive detection of Alzheimer's biomarkers should impact early detection and prognosis

and permit longitudinal studies involving various treatments and their corresponding effects on biomarker levels. One important part of demonstrating the feasibility of our technique is comparing our results to those obtained using an established technique, in this case ELISA. In addition, there is limited literature on the levels of amyloid beta in serum from AD patients.

**Year End Progress Summary:**

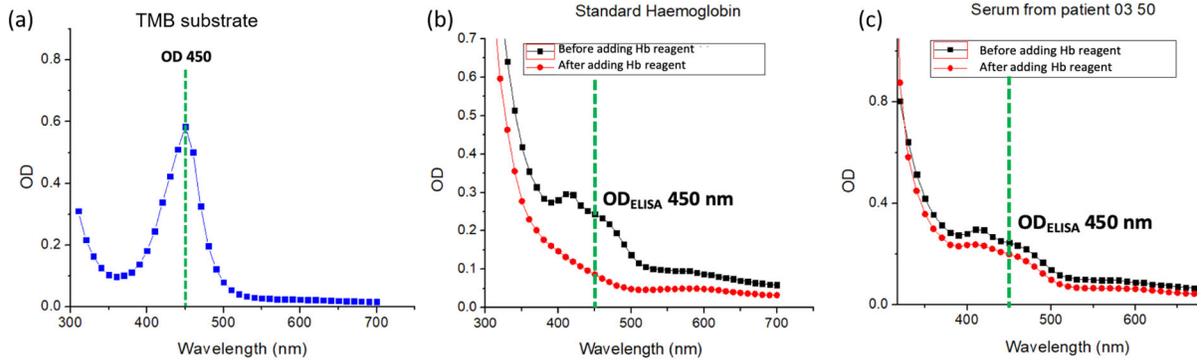


Figure 4. Hemoglobin assay results. This colorimetric assay enables us to measure the amount of hemoglobin present in a sample (a) Optical density (OD) of 3,3',5,5'-Tetramethylbenzidine (TMB) which is the chromogenic substrate used in the assay. (b) OD of a hemoglobin standard before (black) and after (red) adding the hemoglobin detector compound (c) OD of a patient serum sample before (black) and after (red) adding the hemoglobin detector compound.

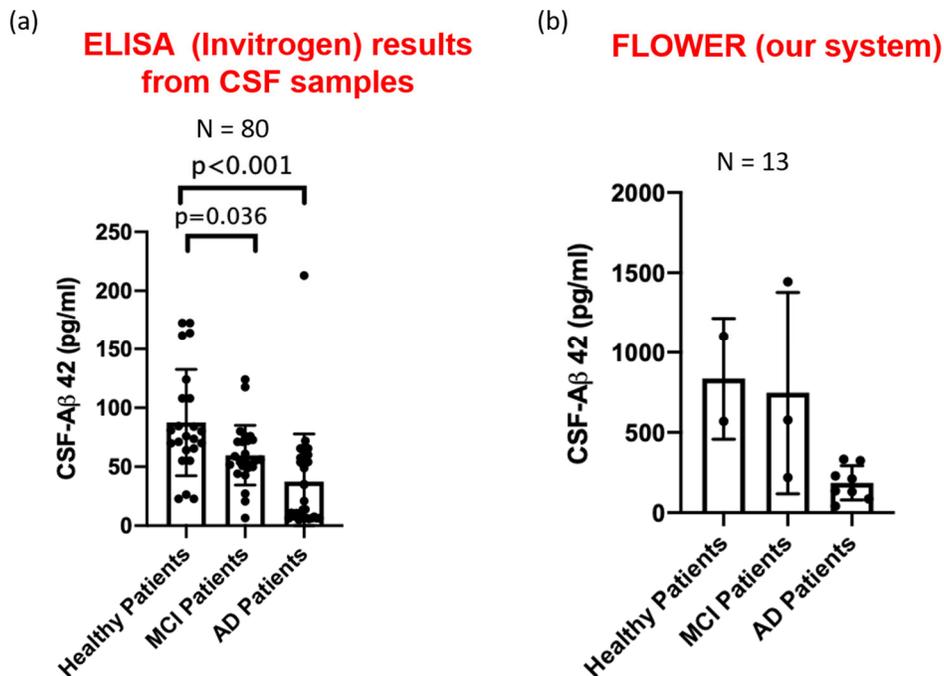


Figure 3. ELISA CSF results. We have screened 80 postmortem patient samples for amyloid-beta 42 using ELISA and have compared this to results obtained using FLOWER. Interestingly, the amyloid beta 42 level in CSF detected by FLOWER's system is higher than was found using ELISA; however, the values found from FLOWER match other values reported in the literature.

As described in Aim 1, we have performed a hemoglobin assay (Figure 2) on our patient samples. Samples with a hemoglobin level of > 0.8 mg/dl were not used as per the kit instructions. We have performed ELISA detection results on 80 CSF samples (Figure 3) and compared them to results we have obtained using FLOWER (Figure 3). 80 instead of 100 patient samples were used as 20 samples had too much hemoglobin in them to be useful. We also performed ELISA on serum postmortem patient samples (Aim 3), however, we were unable to detect any amyloid-beta in the serum. For our next steps, we will try to detect amyloid-beta using FLOWER. As FLOWER has a higher sensitivity than ELISA, we believe we can detect amyloid-beta levels in serum patient samples using FLOWER.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Evaluating the Impact of Carotid Artery Endarterectomy on Cognition and Brain Function using Advanced Neuro-imaging Techniques.** Craig Weinkauf, MD, PhD, Maria Altbach, PhD, Ashley Stokes, PhD, Lee Ryan, PhD, Ted Trouard, PhD. University of Arizona; Barrow Neurological Institute; Arizona Alzheimer's Consortium.

### **Project Description:**

Carotid atherosclerosis is associated with stroke, yet growing evidence links carotid disease to mild cognitive impairment (MCI) and dementia. Medical or surgical intervention is offered to patients with carotid disease for the goal of stroke prevention, but not for prevention or treatment of cognitive dysfunction. In fact, cognition is not regularly evaluated in this patient population and plays no role in clinical decisions.

Using resting-state functional MRI (rs-fMRI) and structural MRI combined with neurocognitive testing, we designed a pilot study to evaluate the brain and cognition in patients who undergo carotid endarterectomy (CEA), which is a surgery to remove blockages in the extracranial carotid arteries. Advanced imaging and cognitive testing was performed pre-operative and 4-6 months post-operative to test the **hypothesis that CEA will result in improved cognition and brain connectivity in patients with significant carotid stenosis.** Our preliminary data are very exciting, yet we need further evaluation of the rs-fMRI data and recruitment of 10 additional control patients for better interpretation of these data and RO1 grant submission.

### **Specific Aims:**

- 1) Recruit 10 control patients who have significant carotid vascular disease, but do not undergo CEA for imaging/testing based on current study parameters.
- 2) Process and evaluate rs-fMRI data of the cohort already imaged and the 10 additional control patients.

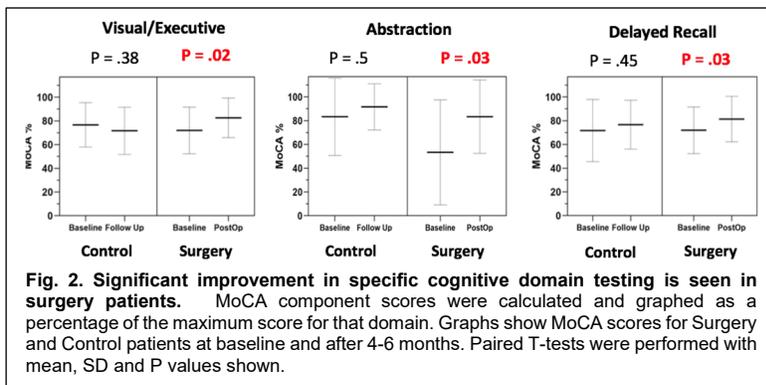
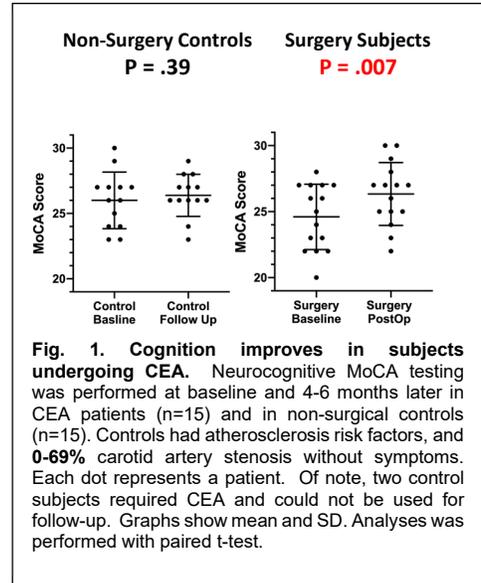
### **Background and Significance:**

Carotid vascular disease is implicated in the cumulative process of vascular cognitive impairment presumably through a combination of impaired blood flow to the brain and subclinical cerebrovascular events<sup>1</sup>. Increasing data indicate carotid atherosclerosis contributes to all forms of dementia including Alzheimer's<sup>2,3</sup>. Although interventions are available to prevent strokes and remove blockages of blood flow to the brain (carotid endarterectomy or stenting), no intervention is directed towards prevention of cognitive decline. Understanding the interplay between carotid disease and dementia/MCI in addition to the effect of interventions to treat carotid vascular disease is particularly valuable because CEA would offer an available intervention to help this patient population.

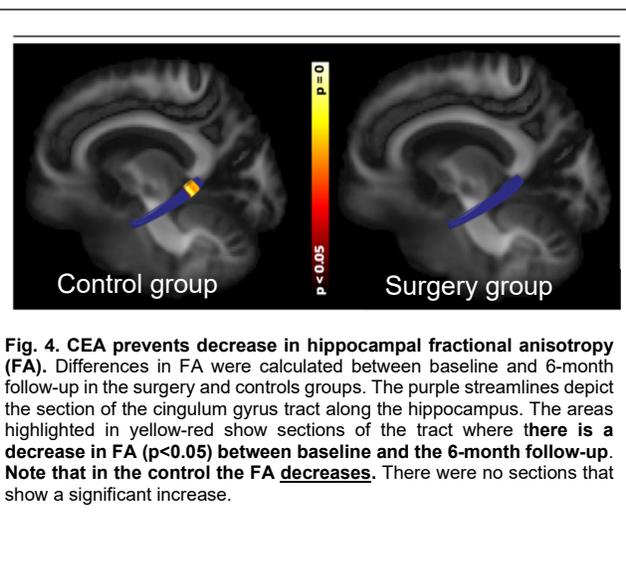
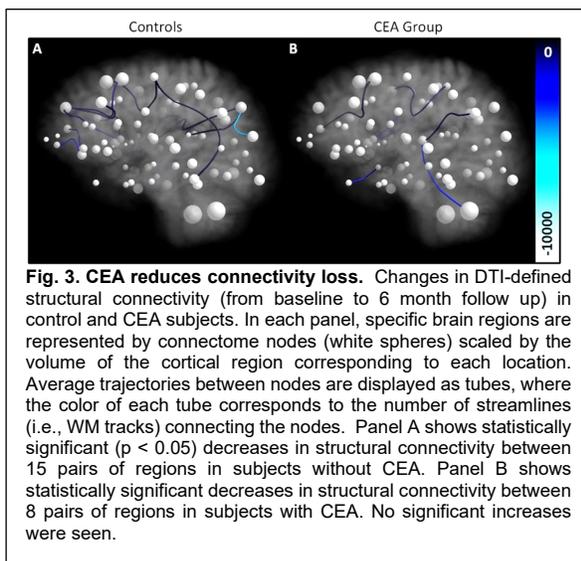
### **Year End Progress Summary:**

Recruitment, testing and analysis of control subjects with ECAD, but who did not undergo CEA was performed. Similar to the intervention subjects (ECAD + CEA) our control subjects (ECAD w/o CEA) had baseline brain imaging and neurocognitive testing performed. In addition, they had 6 month follow-up imaging and neurocognitive testing. We expanded our aims to evaluate key AD-related brain features and cognition in these control subjects compared to intervention subjects over time. Summary of this work includes:

- 1) **rs-fMRI:** after extensive analyses by Dr. Stokes, we have not detected any significant differences in rs-fMRI parameters between baseline and follow-up imaging in our patients with CEA or controls without.
- 2) **Cognitive Evaluation with MoCA:** Our data show that patients undergoing CEA have statistical improvement in MoCA scores at 6 months whereas controls have no significant improvement (**Fig. 1**). In addition, this improvement in cognition is driven by three specific cognitive domains: visual/executive, abstraction and delayed recall (**Fig. 2**).
- 3) **Structural connectivity and fractional anisotropy:** We evaluated baseline and post-operative (or delayed) dwMRI-based structural connectivity data in patients who underwent CEA and controls. **Fig. 3** shows the brain regions with significant changes in structural connectivity. Fifteen brain regions had decreases in connectivity in control patients whereas only 8 regions had decreased connectivity in subjects with CEA. This is in keeping with our hypothesis that



CEA could reduce progression of pathology in this at-risk population. In addition, focusing on the hippocampus because of its relevance in dementia, we found that CEA prevented further statistically significant decrease in FA higher fractional anisotropy (FA), a measure of white matter coherence and structural integrity, compared to control subjects (**Fig. 4**).



**Summary Update:** Overall, we have been successful in subject recruitment. We have continued to not find significant between-cohort or over delayed time differences in rs-fMRI in this patient population. We will continue evaluating these data because this population lends itself well to the possibility of understanding important functional/structural relationships. However, we have found encouraging results that indicate ECAD and its treatment with CEA may be able to modify MCI and key structural brain characteristics, which are indicators of Alzheimer's disease risk.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Effect of ApoE Isoforms on Neuronal- and Astrocytic Energy Metabolism.** Fei Yin, PhD, Haiwei Gu, PhD, Roberta Diaz Brinton, PhD. University of Arizona; Arizona State University; Arizona Alzheimer's Consortium.

### **Specific Aims:**

- 1) To differentiate the mitochondrial phenotype and fuel preference between neurons and astrocytes.
- 2) To determine age- and ApoE isoform-associated changes in neuron and astrocyte energy metabolism.
- 3) To investigate the mechanistic role of ApoE isoforms on neuron-astrocyte metabolic interactions.

### **Background and Significance:**

Brain performs a variety of energy-demanding tasks and thus possesses a high energy consumption rate with most of which generated by mitochondria via oxidative phosphorylation. Moreover, multiple lines of evidence suggest that energy metabolism is key in driving cell proliferation, differentiation, and activation<sup>1-3</sup>. Late-onset Alzheimer's disease (LOAD) has a multifactorial nature and is associated with a decline in brain glucose metabolism and a less efficient mitochondria population. While the ApoE4 effect on brain metabolism is established in late stage AD, mixed results have been reported on its effect in early stages of both pathology- and risk-factor focused AD models<sup>4-6</sup>. We propose that such a discrepancy is relevant to the heterogeneous cellular composition of the brain and the distinct capacity of these cells to metabolize various energy fuels. The mechanism by which the metabolic phenotype of neurons and astrocytes interact with different ApoE isoforms is an under-investigated area of high significance. Determining the process underlying the interactions between different cell types and its shift with disease progression has the potential to identify novel therapeutic target(s) for maintaining or restoring a metabolic homeostasis in the degenerating AD brain.

### **Year End Progress Summary:**

#### **1) Distinctive bioenergetic phenotype and fuel preference between neuron and astrocyte.**

Our data suggest that neurons have a substantial reserve respiratory capacity that equals to its basal respiration while astrocytes have minimum to none reserve capacity in metabolizing glucose. Moreover, astrocytes have more flexibility in metabolizing different energy substrates such as fatty acids through  $\beta$ -oxidation, as supported by elevated expression of key fatty acid metabolism genes that are involved in fatty acid transport, initial  $\beta$ -oxidation and mitochondrial trifunctional protein catalyzed reactions. Consistent with their oxidative nature, neurons are less capable in metabolizing glucose compared to the glycolysis product pyruvate. These data suggest that the distinctive mitochondrial bioenergetic phenotype between neuron and astrocyte can be connected with altered availability and utilization of different energy fuels in the AD brain.

#### **2) ApoE isoforms differentially regulate neuronal- and astrocytic mitochondrial bioenergetic phenotypes.**

To distinguish the effect of ApoE4 on regulating neuronal- and astrocytic mitochondrial function and their preference to different energetic fuels, primary neurons and astrocytes were isolated from the forebrain of humanized ApoE3 and ApoE4 knockin mice. Mitochondrial bioenergetic profile and their dependency and capacity of metabolizing different substrates were characterized.

Both ApoE3 and ApoE4 embryonic neurons relied primarily on glucose-derived fuels than others in terms of both dependency and capacity, whereas astrocytes could metabolize more fatty acids than neurons. Our results further revealed that ApoE4 neurons and astrocytes exhibited lower spare respiration capacity ratio compared to ApoE3 cells, although the basal respiration of ApoE4 astrocytes was higher than that of ApoE3 astrocytes. Across energy fuels, ApoE4 astrocytes had higher maximum capacity metabolizing glucose than fatty acids while such a fuel preference was much less significant in ApoE3 astrocytes. Consistently, the maximal capacity in metabolizing fatty acids was significantly lower in ApoE4 astrocytes when compared to that of ApoE3 astrocytes. Such a reduced capacity to metabolize fatty acids in ApoE4 astrocytes also led to elevated levels of medium to long-chain, fatty acids including palmitic acid, myristic acid and stearic acid relative to ApoE3 controls. Together, our findings suggest that the impact of ApoE genotype on mitochondrial function and brain bioenergetics is cell type dependent, and the development of strategies to restore brain energy metabolism against AD should consider ApoE genotype, disease-stage, and the fuel preference of different cell types in brain.

### **3) ApoE4 Astrocytes Exhibit Diminished Bioenergetic Support to Neurons**

Astrocyte not only provides critical support to neuron for synapse formation/pruning and neurotransmitter modulation, but also participates in the maintenance of metabolic homeostasis in brain. We therefore determined how astrocytes of different ApoE genotypes differentially support neuronal metabolic functions. Metabolically, maximal-, but not basal mitochondrial respiration in ApoE3 neurons was elevated by co-culturing with ApoE3 astrocytes. In parallel, an increase in neuronal glycolysis after the co-culture was evidenced by elevated extracellular acidification rate and increased lactate levels in ApoE3 neurons. Such metabolic enhancements to neurons were significantly diminished in ApoE4 astrocytes when compared to ApoE3 astrocytes. Similar differences between ApoE3 and ApoE4 astrocytes were observed when they were co-cultured with ApoE4 neurons. Hexokinases 1 and 2 (HK1 and HK2) phosphorylate glucose to glucose-6-phosphate and are the rate-limiting enzymes for glycolysis while pyruvate dehydrogenase (PDH) decarboxylate pyruvate to acetyl-CoA for the TCA cycle. In line with the cellular metabolic data, immunoblotting also suggested that co-culture with ApoE3 astrocyte triggered HK1 and HK2 protein expression and reduced inhibitory phosphorylation of PDH in both ApoE3 and ApoE4 neurons, but such effects were diminished when these neurons were co-cultured with ApoE4 astrocytes. Together, these results demonstrate that ApoE4 astrocytes are less capable in supporting neuronal metabolic functions.

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Cognitive Effects of Carotid Intervention-related Brain Microinjury.** Wei Zhou, MD, Ted Trouard, PhD, Ying-hui Chou, PhD, Salil Soman, MD, Greg Zaharchuk, PhD, Chiu-Hsieh Hsu, PhD, Manoj Saranathan, PhD. University of Arizona; Southern Arizona VA Health Care System; Harvard Medical School; Stanford University; Arizona Alzheimer's Consortium.

### **Project Description:**

We have shown that carotid intervention-related micro-embolization affects memory function despite of an absence of neurologic complications<sup>1-3</sup>. However, continuous controversies on the cognitive effects of micro-embolization highlight the critical **GAP** in our understanding of these embolization-related micro-brain injuries. The goal of this pilot project is to characterize cognitive significant microinfarcts (CSMI) using our existing database and to generate additional information on brain function using fMRI. We plan to obtain additional funding in the next 18 months to support our overarching goal of identifying imaging biomarker(s) predictive of cognitive fragility for patients at risk for long-term cognitive deterioration.

### **Specific Aims:**

- 1) To identify CSMI by determining how characteristics of micro-infarcts contribute to cognitive changes in patients undergoing carotid revascularization procedures.
- 2) To validate CSMI by examining brain functional connectivity using fMRI
- 3) (Future) To characterize high risk patterns of brain connectivity contributing to CSMI

### **Background and Significance:**

Carotid atherosclerosis is not only a major contributor of strokes, but also associated with cognitive impairment<sup>4-8</sup>. While carotid artery intervention is an important strategy for stroke prevention, periprocedure subclinical micro-embolization is common, occurring in 30-80% of patients<sup>2, 3, 9, 10</sup> (Fig. 1). Majority of these micro-emboli do not cause clinically evident neurologic sequelae. However, we and others have reported their association with cognitive deterioration<sup>1-3</sup>. Understanding these microembolization-related brain injuries has a significant impact in public health relating to cognitive impairment and risk of dementia in our aging population.

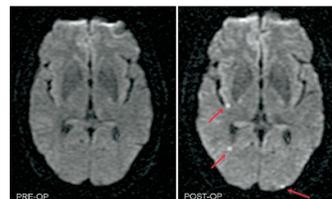


Figure 1: DWI-MRI showed procedure-related microinfarcts

Our central hypothesis is that characteristics of micro-infarcts and baseline brain properties modulate cognitive impacts of procedure-related subclinical embolization. We propose to identify cognitively significant micro-infarcts (CSMI) through a multidisciplinary collaborative team at 4 institutions. By understanding these micro-brain injuries, this project will help to generate information for our long-term goal of understanding cognitive impairment in aging population. The proposal may also change our current clinical practice by identify a subgroup of patients at risk for CSMI and therefore carotid intervention should be restrained in asymptomatic patients.

### **Year End Progress Summary:**

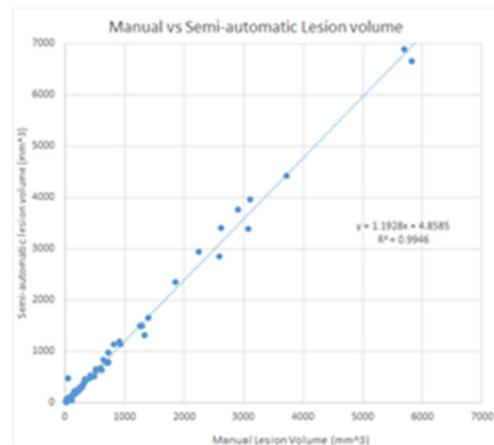
We have made significant progress and have not changed the specific aims

1. To identify CSMI by determining how characteristics of micro-infarcts contribute to cognitive changes in patients undergoing carotid revascularization procedures.
2. To validate CSMI by examining brain functional connectivity using fMRI

1. To identify cognitively significant microinfarcts (CSMI) by determining how characteristics of micro-infarcts contribute to cognitive changes in patients undergoing carotid revascularization procedures.

a. **develop innovative semiautomated software to define lesions.**

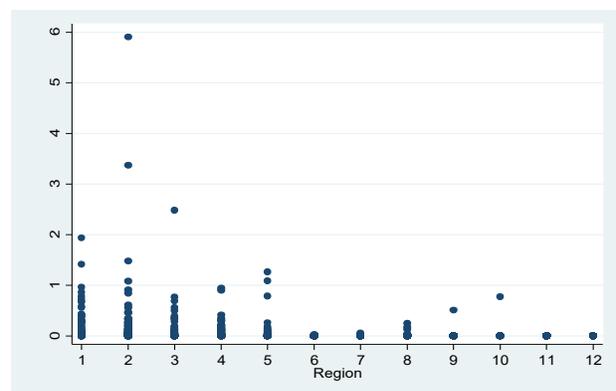
Acute microinfarcts are defined by new hyper-intensities on postoperative DWIs with corresponding hypointense on ADC maps. Postoperative DWIs were compared to the preoperative ones to identify the procedure-related microembolization. To understand carotid intervention-related subclinical embolization, we previously characterized the size of microinfarcts identified on DWI brain images using a manual tracing technique. Through the current collaboration, our investigator team (Trouard Lab) has developed and implemented a semi-automated region-growing registration and algorithm to define size and location of microinfarcts using in-house software. The user selects the center of a lesion and the algorithm identifies the maximum intensity voxel within a user-defined search radius. With this maximum intensity voxel used as a seed, a region is grown by including adjacent voxels with an intensity value within at least a 15% of the maximum value. With the regions of interest (ROI) identified, the program calculates the volume of each lesion using the number of voxels in the segmented regions. This semi-automated approach decreases human variability and operator fatigue. We have validated the program against our previous manually traced lesions and showed good inter-rater and intra-rater reliability (kappa value of 0.32, and intraclass correlation coefficient of 0.995) (Figure A, Unpublished, Manuscript pending).



**Figure A:** Correlation between volumes of SBIs traced manually and calculated using our semi-automated approach

b. **Define location and size of microinfarcts:**

Using the new semi-automated region-growing registration and algorithm program developed by our team, we analyzed our previously recruited 157 subjects from 2012 to 2018. We identified the infarct volume of 540 infarcts in 83 subjects (56.5%) with a mean total infarcts volume of 0.6cm<sup>3</sup> (0 - 6.9cm<sup>3</sup>) related to carotid interventions. In addition, DWIs were skull-stripped using Functional MRI of the Brain (FMRIB) Software Library (FSL)'s BET tool<sup>71</sup>. Following brain extraction, DWIs were registered to the Montreal Neurological Institute (MNI)-152 T1-weighted template provided in the FSL software package<sup>72</sup> using an affine transformation with twelve degrees of freedom as implemented in FSL's FLIRT tool<sup>73</sup>. The estimated affine transformation was then applied to the binary lesion masks generated in the steps described above, such that each lesion mask was also put into the common MNI-152 space. Finally, the locations of lesions were assigned to either the frontal lobe, temporal lobe, parietal lobe, occipital lobe, or other based upon their coordination in the brain using an in-house MATLAB script.



List of the regions

1 Frontal Lobe	2 Parietal Lobe	3 Occipital Lobe	4 Temporal Lobe	5 Misc.	6 Thalamus
7 Caudate	8 Putamen	9 Pallidum	10 Hippocampus	11 Amygdala	12 Accumbens

**Figure B:** Distribution and volume of infarcts within 12 brain regions of the 147 subjects

c. Among 157 subjects with pre- and post- DWIs, 77 subjects (49%) had procedure-related microinfarcts with an average volume of 762.90mm<sup>3</sup> (ranging, 18 to 6892mm<sup>3</sup>). Seven sets of neuropsychological tests focusing on memory, executive function, and language were analyzed in collaboration with Chou's lab and Dr. Hsu, our statistician. Changes in volumes of infarcts were significantly correlated to long-term changes in memory measured by MOANS (P<0.05) and executive function measured by the Trail Making Test (P<0.05). The majority of microinfarcts were located in cortical regions, most commonly parietal and frontal lobes. When the locations of microinfarcts were considered, significant correlations between volumes of microinfarcts and changes in all three cognitive domains were observed (P<0.05) (unpublished, manuscript pending)

2. To validate CSMI by examining brain functional connectivity using fMRI

Recruitment on ongoing. Worked with Drs. Trouard and Chou to standardize the imaging protocol.

**Project Progress Reports**

**University of Arizona**  
**College of Medicine – Phoenix**

# ARIZONA ALZHEIMER'S CONSORTIUM

## 2019-2020 Scientific Progress Report

**Traumatic Brain Injury, Alzheimer's Disease, Probiotics, and Cognitive Function.** Jonathan Lifshitz, PhD, Maha Saber, PhD, M. Luisa Rojas, MS, Daniel R. Griffiths, BS, L. Matthew Law, PhD, Rachel K. Rowe, PhD. Translational Neurotrauma Research Program; UA College of Medicine – Phoenix; Barrow Neurological Institute at Phoenix Children's Hospital; Phoenix VA Health Care System; Arizona Alzheimer's Consortium.

### **Specific Aims:**

1. To interpret neuropathology in a mouse model of Alzheimer's disease in the context of traumatic brain injury in aged mice. *Hypothesis: Similar and discordant neuropathology between 18-month 3xTg-AD (B6;129-Psen1 Tg[APP<sup>Swe</sup>,tau<sup>P301L</sup>]1Lfa) mice without brain injury and wild type mice with diffuse brain injury indicate consequences of acute and chronic disease.* Inflammation will be assessed by myeloid cell population distributions in blood. Physiological disturbance will be assessed by sleep behavior. Neuroinflammation will be assessed by immunohistochemistry and quantified by microglial skeletal and fractal analysis. Additional pathology will include A-beta, Tau, and TDP-43 to capture clinical neuropathological correlates.
2. To evaluate fecal microbiome analysis as a pharmacodynamic outcome in treating diffuse brain injury with a probiotic diet. *Hypothesis: The efficacy of probiotic treatment of brain injury-induced neurological symptoms in the mouse is measurable in fecal microbiome.* Diffuse brain injury by midline fluid percussion induces somatic, cognitive, and emotional impairments. A probiotic of multiple strains (Target GB-X™, Klaire Labs) is provided to treat neurological impairments, with fecal samples collected over the course of disease. Regression analysis can identify relationships between fecal bacteria and neurological impairment and recovery.
3. To evaluate whether *in vivo* cerebrovascular function reflects cognitive function. *Hypothesis: Cerebrovascular imaging and vasoreactivity via in vivo miniscopes encodes functional information related to novel object recognition and spatial navigation that depends on rat age.* Young (3-6 mo), middle (9-12 mo), and aged (22-25 mo) rats will have miniature fluorescent microscopes (miniscopes) secured over the medial prefrontal cortex (mPFC). Intravenous fluorescent dextrans visualize cerebrovasculature while rats participate in novel object recognition tasks and voluntary exploration of the peg forest rehabilitation arena. Real-time blood flow and vessel diameter are collected, synchronized with behavioral video.

### **Background and Significance:**

The bidirectional feedback between peripheral and central inflammation drive pathological signaling in traumatic brain injury (TBI) and Alzheimer's disease (AD). Identifying neuropathology at the intersection of TBI and AD may elucidate shared disease mechanisms. Rather than investigating the additive pathology of TBI and Alzheimer's we elect to pursue shared and divergent physiologies and pathologies to relate disease states to one another.

The vast majority of TBI can be classified as mild, not provoking immediate clinical care, but neurological symptoms can persist or emerge in a delayed manner. Our rodent model of TBI, midline fluid percussion injury, replicates a subset of sensori-motor, cognitive, and emotional symptoms. Practical therapies can aid individuals with TBI to return to their activities of daily living. Nutritional supplements, particularly probiotics, are not FDA regulated and may vary between batches, sources, and labeling. Thus, a reliable, non-invasive pharmacodynamic outcome is

critical to track the therapeutic efficacy of probiotic treatment. We propose that fecal microbiome analysis can serve to track dosing and predict efficacy of probiotic intervention.

Cerebral blood flow increases with neural circuit activity. We can image fluorescent molecules through a cranial window in rodents, such as fluorescent dextrans and nanoparticles to observe cerebral vessel function *in vivo*. For the first time, it is possible to record cerebrovascular function in the awake behaving rodent. Here we propose to record cerebrovascular function in young, middle, and aged rats by quantifying fluorescent dextran movement in vessels of the medial prefrontal cortex (mPFC). If validated, this approach can be applied to study therapeutic interventions for cognitive dysfunction.

### **Year-End Progress Summary:**

Specific Aim 1: All data *in vivo*, flow cytometry, and behavioral data have been collected and analyzed. The results have been prepared for submission to *Frontiers in Neuroscience* with the provisional title “Diffuse brain injury and model of Alzheimer’s disease exhibit disease-specific changes in sleep and peripheral inflammation.” The results showed that TBI and AD had disease-specific increases in peripheral inflammation and sleep profiles. Both brain-injured and 3xTg-AD mice had an overall increase in sleep behavior compared to aged control mice. Of note, the outcomes from these studies supported Bayesian multiple linear regression modeling that was more appropriate to the data structure and advanced our program’s ability to analyze and interpret the data. This study has prepared our team for the complexity of preclinical studies necessary to tackle translational research questions associated with traumatic brain injury and Alzheimer’s disease. All remaining tissue samples have been stored to generate preliminary data for grant application on inflammation and aging. Potential collaborations are being discussed with Dr. Reiman to determine NF-L levels in plasma of brain-injured aged mice and the use of CD11b+Ly6C<sup>high</sup> monocytes in the blood as a peripheral marker for brain injury.

Specific Aim 2: During the past reporting period, the methodology for collecting fecal samples across studies has been optimized. The objective is the regular, non-invasive collection of fecal samples. Challenges were met in terms of improving the throughput and storage of samples, which is now routine in our laboratory. Further, our graduate student has obtained all working protocols from the Cope laboratory at Northern Arizona University. In addition, the Cope laboratory hosted our graduate student for 4 days to gain hands-on expertise with the sample processing, extraction, amplification, and analysis. Following the training, additional supplies and minor equipment was required to minimize contamination and improve yield.

Two studies are underway. The first is the fecal microbiome analysis as part of a larger maternal-TBI project. Due to the elevated incidence rate of physical abuse during pregnancy, studies are underway to evaluate TBI in pregnant mice and in the offspring once they reach maturity. Fecal samples are collected and microbiome analysis ongoing. The second study will establish the inflammatory and microbiome landscape of traumatic brain injury. It becomes necessary to conduct a blood-based inflammatory cell analysis in conjunction with fecal microbiome within our animal facility. From there, the post-injury time course between inflammation and microbiome diversity can be tracked over 10 days post-injury. These studies will initiate and complete in March 2020.

Specific Aim 3: Our work continues to optimize the miniature fluorescent microscope (miniscopes) imaging protocol and analysis. We have successfully implanted GRIN lenses in six rats. All rats survived implantation, and recovered without behavioral deficits. Two rats were eliminated from optimization because of artifacts in the imaging field. Two rats were eliminated from the optimization because the lens was implanted too close to the sagittal sinus, with no blood vessels for analysis in the imaging field. Two rats were deemed suitable to attach a baseplate and image with miniscopes in the peg forest arena. We were successfully able to visualize cortical

blood flow in both rats by marking blood with i.p. or i.v. injection of a fluorescein isothiocyanate–dextran. We recorded data from two rats as they passively explored the peg forest arena. The video data were then analyzed in MATLAB. We analyzed video for changes in vessel diameter and deviations in blood flow. We could determine that the blood vessels analyzed were venous, and therefore unlikely to have a significant change in diameter. We did not see changes in vessel diameter in response to passive exploration. We found that our recording rate was too slow to measure blood velocity. We determined that a video capture rate of >80Hz was necessary to analyze blood flow velocity, and deviations in blood flow. Based on the preliminary analysis, ongoing efforts aim to adjust the field of view to capture more arterioles/capillaries and increase sampling rate. In the interim, new versions of the open source miniscopes have become available. To obtain the necessary images, the miniscopes will be updated from v2 to v4. The updated miniscopes have a larger field of view that will allow us to analyze more blood vessels. The updated miniscopes also have an electronic focus that allows an adjustable imaging plane to optimize the imaging field. The updated miniscopes have a faster sampling rate that can accommodate blood flow velocity and diameter measurements.

### **Proposed One-Year and Long-Term Outcomes:**

The Arizona Alzheimer’s Consortium support for the UA College of Medicine – Phoenix allows the program to expand on TBI related effects on aging, the interaction with peripheral inflammatory systems, and develop novel approaches to evaluate cognitive function. By project end, all data related to the intersection of TBI and AD will be published. These data position Dr. Saber to submit for competitive extramural funding at a new home institution. Our program has obtained expertise in peripheral immune cell quantification as a repeated measure of pathological burden to track disease and therapy. These techniques are applied to ongoing studies.

The fecal microbiome serves as a repeatedly, measurable tissue source to track disease and therapeutic efficacy. Particularly in the case of probiotic and dietary interventions, the fecal microbiome lends insight into the pharmacodynamics of the treatment. Studies are underway to evaluate the time and injury related changes in the fecal microbiome. Towards the close of the funding year, new studies will be initiated to evaluate probiotic interventions. Of particular interest is the relationship between the fecal microbiome and peripheral inflammatory cell populations, which can deliver a comprehensive analysis of the pathophysiology. Next year, these studies will expand to include a clinical pilot project to evaluate the ketogenic diet in severe TBI, using both flow cytometry and the fecal microbiome as pharmacodynamic outcomes. This work constitutes the doctoral dissertation of Ms. Rojas.

The miniscope technology provides unique visualization of the awake-behaving brain. For the first time, we align the miniscopes with cerebrovascular function (flow, diameter) as a surrogate of brain function, akin to the BOLD signal that underlies functional MRI. To this end, ongoing projects incorporate miniscopes with the peg forest arena as a natural exploration task and the novel object cognitive tasks. These techniques continue to be refined and the analysis approaches validated. The intention is to incorporate miniscopes as an outcome measure in upcoming proposals.

**2019 – 2020**  
**Publications, Manuscripts, & Grants**

## 2019 Publications and Manuscripts

Abrahamson EE, Head E, Lott IT, Handen BL, Mufson EJ, Christian BT, Klunk WE, Ikonovic MD. Neuropathological correlates of amyloid PET imaging in Down syndrome. *Dev Neurobiol*. 2019 Jul;79(7):750-766. doi: 10.1002/dneu.22713. Epub 2019 Aug 17. Review. PubMed PMID: 31379087.

Adler CH, Beach TG, Zhang N, Shill HA, Driver-Dunckley E, Caviness JN, Mehta SH, Sabbagh MN, Serrano GE, Sue LI, Belden CM, Powell J, Jacobson SA, Zamrini E, Shprecher D, Davis KJ, Dugger BN, Hentz JG. Unified Staging System for Lewy Body Disorders: Clinicopathologic Correlations and Comparison to Braak Staging. *JNeuropathol Exp Neurol*. 2019 Oct 1;78(10):891-899. doi: 10.1093/jnen/nlz080. PubMed PMID: 31504679.

Adler CH, Serrano GE, Zhang N, Hinni ML, Lott DG, Mehta SH, Sue LI, Intorcchia A, Beach TG. Feasibility of repeat and bilateral submandibular gland needle biopsies in Parkinson's disease. *Parkinsonism Relat Disord*. 2019 Nov;68:69-72. doi: 10.1016/j.parkreldis.2019.10.006. Epub 2019 Oct 7. PubMed PMID: 31621624.

All of Us Research Program Investigators, Denny JC, Rutter JL, Goldstein DB, Philippakis A, Smoller JW, Jenkins G, Dishman E. The "All of Us" Research Program. *N Engl J Med*. 2019 Aug 15;381(7):668-676. doi: 10.1056/NEJMSr1809937. PMID: 31412182

Amofa P, DeFeis B, De Wit L, O'Shea D, Mejia A, Chandler M, Locke D, Fields J, Phatak V, Dean P, Crook J, Smith G. (2019) Functional ability is associated with higher adherence to behavioral interventions in mild cognitive impairment. *The Clinical Neuropsychologist*. [epub Oct 13], 1-19.

Andrews-Hanna JR, Grilli MD, and Irish M (2019) A review and reappraisal of the default network in normal aging and dementia. *Oxford Research Encyclopedia of Psychology*, Oxford University Press, 10.1093/acrefore/9780190236557.013.384

Arboleda-Velasquez JF, Lopera F, O'Hare M, Delgado-Tirado S, Marino C, Chmielewska N, Saez-Torres KL, Amarnani D, Schultz AP, Sperling RA, Leyton-Cifuentes D, Chen K, Baena A, Aguillon D, Rios-Romenets S, Giraldo M, Guzmán-Vélez E, Norton DJ, Pardilla-Delgado E, Artola A, Sanchez JS, Acosta-Urbe J, Lalli M, Kosik KS, Huentelman MJ, Zetterberg H, Blennow K, Reiman RA, Luo J, Chen Y, Thiyyagura P, Su Y, Jun GR, Naymik M, Gai X, Bootwalla M, Ji J, Shen L, Miller JB, Kim LA, Tariot PN, Johnson KA, Reiman EM, Quiroz YT. Resistance to autosomal dominant Alzheimer's disease in an APOE3 Christchurch homozygote: a case report. *Nat Med*. 2019 Nov;25(11):1680-1683. doi: 10.1038/s41591-019-0611-3. Epub 2019 Nov 4. PMID: 31686034

Atri A, Goldfarb D, Sheard S, Shaughnessy L. Current and Emerging Solutions to Challenges in the Management of Alzheimer's Disease. *J Clin Psychiatry*. 2019 Dec 3;80(6). pii: MS18002AH3C. doi: 10.4088/JCP.MS18002AH3C. PubMed PMID: 31846243.

Atri A, Sheard S, Goldfarb D. A Multidisciplinary Approach for Addressing Challenges in Alzheimer's Disease. *J Clin Psychiatry*. 2019 Sep 24;80(5). pii: MS18002WC4C. doi: 10.4088/JCP.MS18002WC4C. PMID: 31556977

Atri A. Current and Future Treatments in Alzheimer's Disease. *Semin Neurol*. 2019 Apr;39(2):227-240. doi: 10.1055/s-0039-1678581. Epub 2019 Mar 29. Review. PubMed PMID: 30925615.

Atri A. The Alzheimer's Disease Clinical Spectrum: Diagnosis and Management. *Med Clin North Am.* 2019 Mar;103(2):263-293. doi: 10.1016/j.mcna.2018.10.009. Review. PubMed PMID: 30704681.

Bacon E, Mishra A, Wang Y, Desai M, Yin F, and Brinton RD (2019) Neuroendocrine aging precedes perimenopause and is regulated by DNA methylation. *Neurobiology of Aging*, 74:213-224.

Balak, C., Benard, M., Schaefer, E., Iqbal, S., Ramsey, K., Ernoult-Lange, M., Mattioli, F., Llaci, L., Geoffroy, V., Courel, M., Naymik, M., Bachman, K.K., Pfundt, R., Rump, P., Ter Beest, J., Wentzensen, I.M., Monaghan, K.G., McWalter, K., Richholt, R., Le Behec, A., Jepsen, W., De Both, M., Belnap, N., Boland, A., Piras, I.S., Deleuze, J.F., Szelinger, S., Dollfus, H., Chelly, J., Muller, J., Campbell, A., Lal, D., Rangasamy, S., Mandel, J.L., Narayanan, V., Huentelman, M., Weil, D. & Piton, A. Rare De Novo Missense Variants in RNA Helicase DDX6 Cause Intellectual Disability and Dismorphic Features and Lead to P-Body Defects and RNA Dysregulation. *Am J Hum Genet* 105, 509-525 (2019).

Ballard C, Atri A, Boneva N, Cummings JL, Frölich L, Molinuevo JL, Tariot PN, Raket LL. Enrichment factors for clinical trials in mild-to-moderate Alzheimer's disease. *Alzheimers Dement (N Y)*. 2019 May 20;5:164-174. doi: 10.1016/j.trci.2019.04.001. eCollection 2019. PMID: 31193334

Berens ME, Sood A, Barnholtz-Sloan JS, Graf JF, Cho S, Kim S, Kiefer J, Byron SA, Halperin RF, Nasser S, Adkins J, Cuyugan L, Devine K, Ostrom Q, Couce M, Wolansky L, McDonough E, Schyberg S, Dinn S, Sloan AE, Prados M, Phillips JJ, Nelson SJ, Liang WS, Al-Kofahi Y, Rusu M, Zavodszky MI, Ginty F (2019) Multiscale, multimodal analysis of tumor heterogeneity in IDH1 mutant vs wild-type diffuse gliomas. *PLoS ONE* 14(12): e0219724; PMID: 31881020.

Bernstein AS, Chen NK, Trouard TP. (2019) Bootstrap analysis of diffusion MRI parameters derived from multiband diffusion MRI. *Magnetic Resonance in Medicine*, 82:1796-1803.

Bhattacharjee S, Lee JK, Patanwala AE, Vadieli N, Malone DC, Knapp SM, Lo-Ciganic WH, Burke WJ. Extent and Predictors of Potentially Inappropriate Antidepressant Use Among Older Adults With Dementia and Major Depressive Disorder *Am J Geriatr Psychiatry*. 2019 Aug;27(8):794-805. doi: 10.1016/j.jagp.2019.02.002. Epub 2019 Feb 7. PMID: 30926273

Brookhouser N, Standage-Beier K, Tekel SJ, Schwarz G, Nguyen T, Wang X, Brafman DA\*. A transient reporter for editing enrichment in human cells. *Nucleic Acids Res.* 2019 Aug 20. pii: gkz713.

Brown, AC, Lavik, E, Stabenfeldt, SE. Biomimetic Strategies to Treat Traumatic Brain Injury by Leveraging Fibrinogen. *Bioconjugate Chemistry*. 2019; 30(7):1951-1956. DOI: 10.1021/acs.bioconjchem.9b00360.

Burke AD, Goldfarb D, Bollam P, Khokher S. Diagnosing and Treating Depression in Patients with Alzheimer's Disease. *Neurol Ther.* 2019 Dec;8(2):325-350. doi: 10.1007/s40120-019-00148-5. Epub 2019 Aug 21. Review. PMID: 31435870

Cardinell, BA, Addington, CP, Stabenfeldt, SE, La Belle, JT. Feasibility of a Traumatic Brain Injury Multi-Analyte Blood Diagnostic in Rat Samples. *Critical Reviews in Biomedical Engineering*. 2019; 47(3): 193-206. DOI: 10.1615/CritRevBiomedEng.2019026108

Carlomagno Y, Chung DC, Yue M, Kurti A, Avendano NM, Castanedes-Casey M, Hinkle KM, Jansen-West K, Daugherty LM, Tong J, Phillips V, Rademakers R, DeTure M, Fryer JD, Dickson DW, Petrucelli L, Cook C. Enhanced phosphorylation of T153 in soluble tau is a defining biochemical feature of the A152T tau risk variant. *Acta Neuropathol Commun.* 2019 Jan 23;7(1):10. doi: 10.1186/s40478-019-0661-2. PMID: 30674342

Chandler MJ, Locke DE, Crook JE, Fields JA, Ball CT, Phatak VS, Dean PM, Morris M, Smith GE. Comparative Effectiveness of Behavioral Interventions on Quality of Life for Older Adults With Mild Cognitive Impairment: A Randomized Clinical Trial. *JAMA Netw Open.* 2019 May 3;2(5):e193016. doi: 10.1001/jamanetworkopen.2019.3016. PubMed PMID: 31099860.

Chauhan G, Adams HHH, Satizabal CL, Bis JC, Teumer A, Sargurupremraj M, Hofer E, Trompet S, Hilal S, Smith AV, Jian X, Malik R, Traylor M, Pulit SL, Amouyel P, Mazoyer B, Zhu YC, Kaffashian S, Schilling S, Beecham GW, Montine TJ, Schellenberg GD, Kjartansson O, Guðnason V, Knopman DS, Griswold ME, Windham BG, Gottesman RF, Mosley TH, Schmidt R, Saba Y, Schmidt H, Takeuchi F, Yamaguchi S, Nabika T, Kato N, Rajan KB, Aggarwal NT, De Jager PL, Evans DA, Psaty BM, Rotter JI, Rice K, Lopez OL, Liao J, Chen C, Cheng CY, Wong TY, Ikram MK, van der Lee SJ, Amin N, Chouraki V, DeStefano AL, Aparicio HJ, Romero JR, Maillard P, DeCarli C, Wardlaw JM, Hernández MDCV, Luciano M, Liewald D, Deary IJ, Starr JM, Bastin ME, Muñoz Maniega S, Slagboom PE, Beekman M, Deelen J, Uh HW, Lemmens R, Brodaty H, Wright MJ, Ames D, Boncoraglio GB, Hopewell JC, Beecham AH, Blanton SH, Wright CB, Sacco RL, Wen W, Thalamuthu A, Armstrong NJ, Chong E, Schofield PR, Kwok JB, van der Grond J, Stott DJ, Ford I, Jukema JW, Vernooij MW, Hofman A, Uitterlinden AG, van der Lugt A, Wittfeld K, Grabe HJ, Hosten N, von Sarnowski B, Völker U, Levi C, Jimenez-Conde J, Sharma P, Sudlow CLM, Rosand J, Woo D, Cole JW, Meschia JF, Slowik A, Thijs V, Lindgren A, Melander O, Grewal RP, Rundek T, Rexrode K, Rothwell PM, Arnett DK, Jern C, Johnson JA, Benavente OR, Wassertheil-Smoller S, Lee JM, Wong Q, Mitchell BD, Rich SS, McArdle PF, Geerlings MI, van der Graaf Y, de Bakker PIW, Asselbergs FW, Srikanth V, Thomson R, McWhirter R, Moran C, Callisaya M, Phan T, Rutten-Jacobs LCA, Bevan S, Tzourio C, Mather KA, Sachdev PS, van Duijn CM, Worrall BB, Dichgans M, Kittner SJ, Markus HS, Ikram MA, Fornage M, Launer LJ, Seshadri S, Longstreth WT Jr, Debette S; Stroke Genetics Network (SiGN), the International Stroke Genetics Consortium (ISGC), METASTROKE, Alzheimer's Disease Genetics Consortium (ADGC), and the Neurology Working Group of the Cohorts for Heart and Aging Research in Genomic Epidemiology (CHARGE) Consortium. Genetic and lifestyle risk factors for MRI-defined brain infarcts in a population-based setting. *Neurology.* 2019 Jan 16. PMID: 30651383

Chen L, Li C, Liu Y, Su, J, McLeod, E (2019) Simulating robust far-field coupling to traveling waves in large three-dimensional nanostructured high-Q microresonators, *Photonics Research*, 7:967-976.

Chen NK, Wu PH (2019) The use of Fourier-domain analyses for unwrapping phase images of low signal-to-noise ratio. *Magnetic Resonance in Medicine*, 82:356-366.

Chen Y, Lv C, Li X, Zhang J, Chen K, Liu Z, Li H, Fan J, Qin T, Luo L, Zhang Z. The positive impacts of early-life education on cognition, leisure activity, and brain structure in healthy aging. *Aging (Albany NY).* 2019 Jul 17;11(14):4923-4942. doi: 10.18632/aging.102088. PMID: 31315089

Chen Y, Qi D, Qin T, Chen K, Ai M, Li X, Li H, Zhang J, Mao H, Yang Y, Zhang Z. Brain Network Connectivity Mediates Education-related Cognitive Performance in Healthy Elderly Adults. *Curr Alzheimer Res.* 2019;16(1):19-28. PMID: 30345918

Chew J, Cook C, Gendron TF, Jansen-West K, Del Rosso G, Daugherty LM, Castanedes-Casey M, Kurti A, Stankowski JN, Disney MD, Rothstein JD, Dickson DW, Fryer JD, Zhang YJ, Petrucelli L. Aberrant deposition of stress granule-resident proteins linked to C9orf72-associated TDP-43 proteinopathy. *Mol Neurodegener.* 2019 Feb 15;14(1):9. PMID: 30767771

Chiu MJ, Lue LF, Sabbagh MN, Chen TF, Chen HH, Yang SY. Long-Term Storage Effects on Stability of A $\beta$ (1-40), A $\beta$ (1-42), and Total Tau Proteins in Human Plasma Samples Measured with Immunomagnetic Reduction Assays. *Dement Geriatr Cogn Dis Extra.* 2019 Feb 12;9(1):77-86. doi: 10.1159/000496099. eCollection 2019 Jan-Apr. PubMed PMID: 31043966.

Chou Y-H, Ton That V, and Sundman M (2019) A systematic review and meta-analysis of rTMS effects on cognitive enhancement in mild cognitive impairment and Alzheimer's disease. *Neurobiology of Aging*, 86:1-20, doi: 10.1016/j.neurobiolaging.2019.08.020.

Christensen KR, Beach TG, Serrano GE, Kanaan NM. Pathogenic tau modifications occur in axons before the somatodendritic compartment in mossy fiber and Schaffer collateral pathways. *Acta Neuropathol Commun.* 2019 Feb 28;7(1):29. doi: 10.1186/s40478-019-0675-9. PubMed PMID: 30819250.

Cicognola C, Brinkmalm G, Wahlgren J, Portelius E, Gobom J, Cullen NC, Hansson O, Parnetti L, Constantinescu R, Wildsmith K, Chen HH, Beach TG, Lashley T, Zetterberg H, Blennow K, Höglund K. Novel tau fragments in cerebrospinal fluid: relation to tangle pathology and cognitive decline in Alzheimer's disease. *Acta Neuropathol.* 2019 Feb;137(2):279-296. doi: 10.1007/s00401-018-1948-2. Epub 2018 Dec 13. PubMed PMID: 30547227.

Cui JZ, Lee L, Sheng X, Chu F, Gibson CP, Aydinian T, Walker DC, Sandor GGS, Bernatchez P, Tibbits GF, van Breemen C, Esfandiarei M. In vivo characterization of doxycycline-mediated protection of aortic function and structure in a mouse model of Marfan syndrome-associated aortic aneurysm. *Sci Rep.* 2019 Feb 14; 9(1):2071.

Das, S., Extracellular, R.N.A.C.C., Ansel, K.M., Bitzer, M., Breakefield, X.O., Charest, A., Galas, D.J., Gerstein, M.B., Gupta, M., Milosavljevic, A., McManus, M.T., Patel, T., Raffai, R.L., Rozowsky, J., Roth, M.E., Saugstad, J.A., Van Keuren-Jensen, K., Weaver, A.M. & Laurent, L.C. The Extracellular RNA Communication Consortium: Establishing Foundational Knowledge and Technologies for Extracellular RNA Research. *Cell* 177, 231-242 (2019).

De Wit L, Chandler M, Amofa P, DeFeis B, Mejia A, O'Shea D, Locke D, Fields J, Smith G. 2019. Memory Support System training in mild cognitive impairment: Predictors of learning and adherence. *Neuropsychology Rehabilitation.* [Epub Sept 20], 1-13 doi: 10.1080/09602011.2019.1667833. PubMed PMID: 31538854.

Deeb W, Salvato B, Almeida L, Foote KD, Amaral R, Germann J, Rosenberg PB, Tang-Wai DF, Wolk DA, Burke AD, Salloway S, Sabbagh MN, Chakravarty MM, Smith GS, Lyketsos CG, Lozano AM, Okun MS. Fornix-Region Deep Brain Stimulation-Induced Memory Flashbacks in Alzheimer's Disease. *N Engl J Med.* 2019 Aug 22;381(8):783-785. doi: 10.1056/NEJMc1905240. PubMed PMID: 31433930.

Desai MK and Brinton RD (2019) Autoimmune disease in women: Endocrine transition and risk across the lifespan. *Front Endocrinol (Lausanne).* doi:10.3389/fendo.2019.00265. eCollection 2019.

Dey KK, Wang H, Niu M, Bai B, Wang X, Li Y, Cho JH, Tan H, Mishra A, High AA, Chen PC, Wu Z, Beach TG, Peng J. Deep undepleted human serum proteome profiling toward biomarker discovery for Alzheimer's disease. *Clin Proteomics*. 2019 Apr 17;16:16. doi: 10.1186/s12014-019-9237-1. eCollection 2019. PubMed PMID: 31019427.

Dong Q, Zhang W, Wu J, Li B, Schron EH, McMahon T, Shi J, Gutman BA, Chen K, Baxter LC, Thompson PM, Reiman EM, Caselli RJ, Wang Y. Applying surface-based hippocampal morphometry to study APOE-E4 allele dose effects in cognitively unimpaired subjects. *Neuroimage Clin*. 2019;22:101744. doi:10.1016/j.nicl.2019.101744. Epub 2019 Mar 4. PubMed PMID: 30852398; PubMed Central PMCID: PMC6411498.

Dong Q, Zhang W, Wu J, Li B, Schron EH, McMahon T, Shi J, Gutman BA, Chen K, Baxter LC, Thompson PM, Reiman EM, Caselli RJ, Wang Y. Applying Surface-Based Hippocampal Morphometry to Study APOE-e4 Allele Dose Effects in Cognitively Unimpaired Subjects. *NeuroImage: Clinical*, 2019, 22:101744.

Dong X, Liao Z, Gritsch D, Hadzhiev Y, Bai Y, Locascio JJ, Guennewig B, Liu G, Blauwendraat C, Wang T, Adler CH, Hedreen JC, Faull RLM, Frosch MP, Nelson PT, Rizzu P, Cooper AA, Heutink P, Beach TG, Mattick JS, Müller F, Scherzer CR. Publisher Correction: Enhancers active in dopamine neurons are a primary link between genetic variation and neuropsychiatric disease. *Nat Neurosci*. 2019 Jan;22(1):144-147. doi: 10.1038/s41593-018-0277-z. PubMed PMID: 30482917.

Driver-Dunckley ED, Zhang N, Adler CH, Serrano GE, Sue LI, Shill HA, Mehta SH, Belden CM, Zamrini EY, Davis K, Beach TG. Brain Lewy-Type Synucleinopathy Density Is Associated with a Lower Prevalence of Atherosclerotic Cardiovascular Disease Risk Factors in Patients with Parkinson's Disease. *J Parkinsons Dis*. 2019;9(3):543-552. doi: 10.3233/JPD-191610. PubMed PMID: 31282425.

Driver-Dunckley ED, Zhang N, Shill HA, Mehta SH, Belden CM, Zamrini EY, Davis K, Beach TG, Adler CH. Correlation Between the Movement Disorder Society's Unified Parkinson's Disease Rating Scale and Nonmotor Scales in Patients with Parkinson's Disease. *Innov Clin Neurosci*. 2019 Sep 1;16(9-10):27-29. PubMed PMID:32082946.

Dugger BN, Hoffman BR, Scroggins A, Serrano GE, Adler CH, Shill HA, Belden CM, Sabbagh MN, Caviness JN, Driver Dunckley E, Beach TG. Tau immunoreactivity in peripheral tissues of human aging and select tauopathies. *Neurosci Lett*. 2019 Mar 23;696:132-139. doi: 10.1016/j.neulet.2018.12.031. Epub 2018 Dec 21. PubMed PMID: 30579993.

Dumitrescu, L., Barnes, L.L., Thambisetty, M., Beecham, G., Kunkle, B., Bush, W.S., Gifford, K.A., Chibnik, L.B., Mukherjee, S., De Jager, P.L., Kukull, W., Crane, P.K., Resnick, S.M., Keene, C.D., Montine, T.J., Schellenberg, G.D., Deming, Y., Chao, M.J., Huentelman, M., Martin, E.R., Hamilton-Nelson, K., Shaw, L.M., Trojanowski, J.Q., Peskind, E.R., Cruchaga, C., Pericak-Vance, M.A., Goate, A.M., Cox, N.J., Haines, J.L., Zetterberg, H., Blennow, K., Larson, E.B., Johnson, S.C., Albert, M., Alzheimer's Disease Genetics, C., the Alzheimer's Disease Neuroimaging, I., Bennett, D.A., Schneider, J.A., Jefferson, A.L. & Hohman, T.J. Sex differences in the genetic predictors of Alzheimer's pathology. *Brain* 142, 2581-2589 (2019).

Dunn KE, Huhn AS, Bergeria CL, Gipson CD, Weerts EM. Non-Opioid Neurotransmitter Systems that Contribute to the Opioid Withdrawal Syndrome: A Review of Preclinical and Human Evidence.

J Pharmacol Exp Ther. 2019 Nov;371(2):422-452. doi: 10.1124/jpet.119.258004. Epub 2019 Aug 7. Review. PubMed PMID: 31391211; PubMed Central PMCID: PMC6863456

Ebbert MTW, Jensen TD, Jansen-West K, Sens JP, Reddy JS, Ridge PG, Kauwe JSK, Belzil V, Pregent L, Carrasquillo MM, Keene D, Larson E, Crane P, Asmann YW, Ertekin-Taner N, Younkin SG, Ross OA, Rademakers R, Petrucelli L, Fryer JD. Systematic analysis of dark and camouflaged genes reveals disease-relevant genes hiding in plain sight. *Genome Biol.* 2019 May 20;20(1):97. doi: 10.1186/s13059-019-1707-2. PMID: 31104630

Egan MF, Kost J, Voss T, Mukai Y, Aisen PS, Cummings JL, Tariot PN, Vellas B, van Dyck CH, Boada M, Zhang Y, Li W, Furtek C, Mahoney E, Harper Mozley L, Mo Y, Sur C, Michelson D. Randomized Trial of Verubecestat for Prodromal Alzheimer's Disease. *N Engl J Med.* 2019 Apr 11;380(15):1408-1420. doi: 10.1056/NEJMoa1812840. PMID: 30970186

Egan MF, Mukai Y, Voss T, Kost J, Stone J, Furtek C, Mahoney E, Cummings JL, Tariot PN, Aisen PS, Vellas B, Lines C, Michelson D. Further analyses of the safety of verubecestat in the phase 3 EPOCH trial of mild-to-moderate Alzheimer's disease. *Alzheimers Res Ther.* 2019 Aug 7;11(1):68. doi: 10.1186/s13195-019-0520-1. PMID: 31387606

Ehsani H, Mohler MJ, O'Connor K, Zamrini E, Tirambulo C, Toosizadeh N. The association between cognition and dual-tasking among older adults: the effect of motor function type and cognition task difficulty. *Clin Interv Aging.* 2019 Apr 8;14:659-669. doi: 10.2147/CIA.S198697. eCollection 2019. PubMed PMID: 31040655.

El-Hayek YH, Wiley RE, Khoury CP, Daya RP, Ballard C, Evans AR, Karran M, Molinuevo JL, Norton M, Atri A. Tip of the Iceberg: Assessing the Global Socioeconomic Costs of Alzheimer's Disease and Related Dementias and Strategic Implications for Stakeholders. *J Alzheimers Dis.* 2019;70(2):323-341. doi: 10.3233/JAD-190426. Review. PubMed PMID: 31256142.

Ellis, TW Jr, S Brownstein, K Beitchman, J Lifshitz. (2019) Restoring More Than Smiles in Broken Homes: Dental and Oral Biomarkers of Brain Injury in Domestic Violence. *Journal of Aggression, Maltreatment & Trauma* 28(7): 838-84

El-Nazer R, Adler CH, Beach TG, Belden CM, Artz J, Shill HA, Driver-Dunckley E, Mehta SH, Sabbagh MN, Serrano GE, Sue LI, Zamrini E, Benge JF. Regional neuropathology distribution and verbal fluency impairments in Parkinson's disease. *Parkinsonism Relat Disord.* 2019 Aug;65:73-78. doi:10.1016/j.parkreldis.2019.05.014. Epub 2019 May 10. PubMed PMID: 31109728.

Enriquez D, Elyaderani A, Antone J, Geiger P, Adkins J, Serrano GE, Mastroeni, D, Beach TG, Readhead B, Dudley J. 2019. Single nuclei and single whole cell sequencing of Alzheimer's disease. *Alzheimer's & Dementia: The Journal of the Alzheimer's Association*, 15, P1503.

Escott-Price, V., Baker, E., Shoai, M., Leonenko, G., Myers, A.J., Huentelman, M. & Hardy, J. Genetic analysis suggests high misassignment rates in clinical Alzheimer's cases and controls. *Neurobiol Aging* 77, 178-182 (2019).

Esfandiarei M, Hoxha B, Talley NA, Anderson MR, Alkhouli MF, Squire MA, Eckman DM, Babu JR, Loapschuk GD, Broderick TL. Beneficial effects of resveratrol and exercise training on cardiac and aortic function and structure in the 3xTg mouse model of Alzheimer's disease. *Drug Des Devel Ther.* 2019 Apr 17; 13:1197-1211.

Espeland MA, Hayden KM, Lockhart SN, Yassine HN, Hoscheidt S, Yasar S, Luchsinge, JA, Neiberg RH, Brinton RD, Charmichael O (2019) Action for Health in Diabetes Brain Magnetic Resonance Imaging (Look AHEAD Brain) Research Group, Sex--related differences in brain volumes and cerebral blood flow among overweight and obese adults with type 2 diabetes: Exploratory analyses from the action for health in diabetes brain magnetic resonance imaging study. *J Gerontol A Biol Sci Med Sci*, doi:10.1093/gerona/glz090. [Epub ahead of print]

Feng, M, Kim J, Field K, Reid C, Chatzistamou I, and Shim M. (2019) Aspirin ameliorates the long-term adverse effects of doxorubicin through suppression of cellular senescence *FASEB BioAdvances* 1: 579-590

Fernandez F (2019) Circadian responses to fragmented light: Research synopsis in humans. *Yale Journal of Biology & Medicine*, 92:337-348.

Fernandez HH, Stamler D, Davis MD, Factor SA, Hauser RA, Jimenez-Shahed J, Ondo WG, Jarskog LF, Woods SW, Bega D, LeDoux MS, Shprecher DR, Anderson KE (2019). Long-term safety and efficacy of deutetrabenazine for the treatment of tardive dyskinesia. *J Neurol Neurosurg Psychiatry*, 90(12), 1317-1323.

Fonseca-Portilla R, Krell-Roesch J, Shaibi GQ, Caselli RJ, Mandarino LJ, Zhang N, Hentz JG, Coletta DK, de Filippis EA, Dawit S, Geda YE. Brain-Derived Neurotrophic Factor and Its Associations with Metabolism and Physical Activity in a Latino Sample. *Metab Syndr Relat Disord*. 2019 Mar; 17 (2):75-80 Epub 2018 Nov 10 PMID: 30418087 PMCID: 6484340 DOI: 10.1089/met.2018.0028

Foster N, Bondi M, Das R, Foss M, Hershey L, Koh S, Logan R, Poole C, Shega J, Sood A, Thothala N, Wicklund M, Yu M, Bennett A, Wang D. Quality Improvement in Neurology: Mild Cognitive Impairment Measurement Set. *Neurology* 2019 Oct 15;93(16):705-713.

Fox KC, Andrews-Hanna JR, Mills C, Dixon ML, Markovic J, Thompson E, Irving Z, Christoff K Affective neuroscience of self-generated thought. *Annals of the New York Academy of Sciences*, doi: 10.1111/nyas.13740. [Epub ahead of print].

Francois-Moutal L, Felemban R, Scott RD, Sayegh MR, Miranda VG, Perez-Miller, S, Khanna R, Gokhale V, Zarnescu DC, Khanna M (2019) Small molecular targeting TDP-43's RNA recognition motifs reduces locomotor defects in a drosophila model of amyotrophic lateral sclerosis (ALS). *ACS Chemical Biology*, 14:2006-2013.

Franzmeier N, Rubinski A, Neitzel J, Ewers M; Alzheimer's Disease Neuroimaging Initiative (ADNI). The BIN1 rs744373 SNP is associated with increased tau-PET levels and impaired memory. *Nat Commun*. 2019 Apr 16;10(1):1766. doi: 10.1038/s41467-019-09564-5. PMID: 30992433

Frölich L, Atri A, Ballard C, Tariot PN, Molinuevo JL, Boneva N, Geist MA, Raket LL, Cummings JL. Open-Label, Multicenter, Phase III Extension Study of Idalopirdine as Adjunctive to Donepezil for the Treatment of Mild-Moderate Alzheimer's Disease. *J Alzheimers Dis*. 2019;67(1):303-313. doi: 10.3233/JAD-180595. PMID: 30636738

Frow EK, Brafman DA, Muldoon A, Krum L, Williams P, Becker B, Nelson JP, Pritchett A. Characterizing direct-to-consumer stem cell businesses in the southwest United States. *Stem Cell Reports*. 2019 Aug 13;13(2):247-253.

Fu H, Possenti A, Freer R, Nakano Y, Hernandez Villegas NC, Tang M, Cauhy PVM, Lassus BA, Chen S, Fowler SL, Figueroa HY, Huey ED, Johnson GVW, Vendruscolo M, Duff KE. A tau homeostasis signature is linked with the cellular and regional vulnerability of excitatory neurons to tau pathology. *Nat Neurosci.* 2019 Jan;22(1):47-56. doi: 10.1038/s41593-018-0298-7. Epub 2018 Dec 17.

Fuller JT, Cronin-Golomb A, Gatchel JR, Norton DJ, Guzmán-Vélez E, Jacobs HIL, Hanseeuw B, Paredilla-Delgado E, Artola A, Baena A, Bocanegra Y, Kosik KS, Chen K, Tariot PN, Johnson K, Sperling RA, Reiman EM, Lopera F, Quiroz YT. Biological and Cognitive Markers of Presenilin1 E280A Autosomal Dominant Alzheimer's Disease: A Comprehensive Review of the Colombian Kindred. *J Prev Alzheimers Dis.* 2019;6(2):112-120. doi: 10.14283/jpad.2019.6. PMID: 30756118

Gao S, Chen Y, Sang F, Yang Y, Xia J, Li X, Zhang J, Chen K, Zhang Z. White Matter Microstructural Change Contributes to Worse Cognitive Function in Patients With Type 2 Diabetes. *Diabetes.* 2019 Nov;68(11):2085-2094. doi: 10.2337/db19-0233. Epub 2019 Aug 22. PMID: 31439643

Garcia A, Natri Hp, Buetow KH, Trumble B and Wilson MA. 2019. Evolution of immune sexual dimorphism in response to placental invasiveness. *Trends in Genetics* 36(1):5-7. doi: 10.1016/j.tig.2019.10.007

Gardner HL, Sivaprakasam K, Briones N, Zismann V, Perdignes N, Drenner K, Facista S, Richholt R, Liang W, Aldrich J, Trent JM, Shields P, Robinson N, Johnson J, Lana S, Houghton P, Fenger J, Lorch G, Janeway KA, London CA\*, Hendricks WPD\* (2019) Canine osteosarcoma genome sequencing identified recurrent mutations in *DMD* and the histone methyltransferase gene *SETD2* *Communications Biology* Jul 19; 2:266; PMID: 31341965.

Gaynor L, Curiel R, Penate A, Roselli M, Burke S, Wicklund M, Loewenstein D, and Bauer R. Visual Object Discrimination as an Early Predictor of Mild Cognitive Impairment and Alzheimer's Disease. *J Int Neuropsychol Soc* May 2019; 21: 1-11.

Ginsberg SD, Malek-Ahmadi MH, Alldred MJ, Che S, Elarova I, Chen Y, Jeanneteau F, Kranz TM, Chao MV, Counts SE, Mufson EJ. Selective decline of neurotrophin and neurotrophin receptor genes within CA1 pyramidal neurons and hippocampus proper: Correlation with cognitive performance and neuropathology in mild cognitive impairment and Alzheimer's disease. *Hippocampus.* 2019 May;29(5):422-439. doi: 10.1002/hipo.22802. Epub 2017 Sep 27. PubMed PMID: 28888073

Ginsberg SD, Malek-Ahmadi MH, Alldred MJ, Chen Y, Chen K, Chao MV, Counts SE, Mufson EJ. Brain-derived neurotrophic factor (BDNF) and TrkB hippocampal gene expression are putative predictors of neuritic plaque and neurofibrillary tangle pathology. *Neurobiol Dis.* 2019 Dec;132:104540. doi: 10.1016/j.nbd.2019.104540. Epub 2019 Jul 23. PubMed PMID: 31349032.

Ginsberg, S. D., Malek-Ahmadi, M., Alldred, M. J., Che, S., Elarova, I., Chen, Y., Jeanneteau, F., Kranz, T. M., Chao, M. V., Counts, S E. and E. J. Mufson: Selective decline of neurotrophin and neurotrophin receptor genes within CA1 pyramidal neurons and hippocampus proper: Correlation with cognitive decline and neuropathology in mild cognitive impairment and Alzheimer's disease, *Hippocampus*, 29:422-439, 2019. PMID: 28888073. PMCID: PMC5844851. doi: 10.1002/hipo.22802.

Goenaga J., Powell G.L., Leyrer-Jackson J.M., Piña J., Phan S., Prakapenka A.V., Koebele S.V., Namba M.D., McClure E.A., Bimonte-Nelson H.A., Gipson C.D. (2019). N-acetylcysteine yields sex-specific efficacy for cue-induced reinstatement of nicotine seeking. *Addict Biol.* doi: 10.1111/adb.12711. PMID: 30734439

Goldfarb D, Sheard S, Shaughnessy L, Atri A. Disclosure of Alzheimer's Disease and Dementia: Patient- and Care Partner-Centric Decision-Making and Communication. *J Clin Psychiatry.* 2019 Mar 19;80(2). pii: MS18002BR1C. doi: 10.4088/JCP.MS18002BR1C. PubMed PMID: 30900850.

Gordon BA, Blazey TM, Christensen J, Dincer A, Flores S, Keefe S, Chen C, Su Y, McDade EM, Wang G, Li Y, Hassenstab J, Aschenbrenner A, Hornbeck R, Jack CR, Ances BM, Berman SB, Brosch JR, Galasko D, Gauthier S, Lah JJ, Masellis M, van Dyck CH, Mintun MA, Klein G, Ristic S, Cairns NJ, Marcus DS, Xiong C, Holtzman DM, Raichle ME, Morris JC, Bateman RJ, Benzinger TLS. Tau PET in autosomal dominant Alzheimer's disease: relationship with cognition, dementia and other biomarkers. *Brain.* 2019 Apr 1;142(4):1063-1076. doi: 10.1093/brain/awz019. PMID: 30753379

Graf A, Risson V, Gustavsson A, Bezlyak V, Caputo A, Tariot PN, Langbaum JB, Lopez Lopez C, Viglietta V. Assessment of Clinical Meaningfulness of Endpoints in the Generation Program by the Insights to Model Alzheimer's Progression in Real Life (iMAP) Study. *J Prev Alzheimers Dis.* 2019;6(2):85-89. doi: 10.14283/jpad.2018.49. PMID: 30756114

Grandner M, Khader W, Warlick C, and Fernandez F (2019) Acculturation and Sleep: Implications for Sleep and Health Disparities. *Sleep*, 42:1-2.

Gray DT, and Barnes CA (2019) Experiments in macaque monkeys provide critical insights into age-associated changes in cognitive and sensory function. *Proceedings of the National Academy of Sciences*, 116:26247-26254.

Grilli MD, Coste S, Landry J, and Mangen K (2019) Evidence that an episodic mode of thinking facilitates encoding of perceptually rich memories for naturalistic events relative to a gist-based mode of thinking. *Memory*, 27:1468-1474.

Grossberg GT, Tong G, Burke AD, Tariot PN. Present Algorithms and Future Treatments for Alzheimer's Disease. *J Alzheimers Dis.* 2019;67(4):1157-1171. doi: 10.3233/JAD-180903. Review. PMID: 30741683

Guerreiro R, Escott-Price V, Hernandez DG, Kun-Rodrigues C, Ross OA, Orme T, Neto JL, Carmona S, Dehghani N, Eicher JD, Shepherd C, Parkkinen L, Darwent L, Heckman MG, Scholz SW, Troncoso JC, Pletnikova O, Dawson T, Rosenthal L, Ansorge O, Clarimon J, Lleo A, Morenas-Rodriguez E, Clark L, Honig LS, Marder K, Lemstra A, Rogaeva E, St George-Hyslop P, Londos E, Zetterberg H, Barber I, Braae A, Brown K, Morgan K, Troakes C, Al-Sarraj S, Lashley T, Holton J, Compta Y, Van Deerlin V, Serrano GE, Beach TG, Lesage S, Galasko D, Masliah E, Santana I, Pastor P, Diez-Fairen M, Aguilar M, Tienari PJ, Myllykangas L, Oinas M, Revesz T, Lees A, Boeve BF, Petersen RC, Ferman TJ, Graff-Radford N, Cairns NJ, Morris JC, Pickering-Brown S, Mann D, Halliday GM, Hardy J, Trojanowski JQ, Dickson DW, Singleton A; International Parkinson's Disease Genomics Consortium, Stone DJ, Bras J. Heritability and genetic variance of dementia with Lewy bodies. *Neurobiol Dis.* 2019 Jul;127:492-501. doi: 10.1016/j.nbd.2019.04.004. Epub 2019 Apr 3. PubMed PMID: 30953760.

Gupta HV, Zhang N, Driver-Dunckley E, Mehta SH, Beach TG, Adler CH. Contrast Acuity With Different Colors in Parkinson's Disease. *Mov Disord Clin Pract*. 2019 Sep 6;6(8):672-677. doi: 10.1002/mdc3.12821. eCollection 2019 Nov. PubMed PMID: 31745477.

Gupta Y, Lama RK, Kwon GR; Alzheimer's Disease Neuroimaging Initiative. Prediction and Classification of Alzheimer's Disease Based on Combined Features From Apolipoprotein-E Genotype, Cerebrospinal Fluid, MR, and FDG-PET Imaging Biomarkers. *Front Comput Neurosci*. 2019 Oct 16;13:72. doi: 10.3389/fncom.2019.00072. eCollection 2019. PMID: 31680923

Hanseeuw BJ, Lopera F, Sperling RA, Norton DJ, Guzman-Velez E, Baena A, Paredilla-Delgado E, Schultz AP, Gatchel J, Jin D, Chen K, Reiman EM, Johnson KA, Quiroz YT. Striatal amyloid is associated with tauopathy and memory decline in familial Alzheimer's disease. *Alzheimers Res Ther*. 2019 Feb 4;11(1):17. doi: 10.1186/s13195-019-0468-1. PMID: 30717814

Haure-Mirande, JV, Wang M, Audrain M, Fanutza T, Kim SH, Heja S, Readhead B, Dudley JT, Blitzer RD, Schadt EE. 2019. Integrative approach to sporadic Alzheimer's disease: deficiency of TYROBP in cerebral A $\beta$  amyloidosis mouse normalizes clinical phenotype and complement subnetwork molecular pathology without reducing A $\beta$  burden. *Molecular psychiatry*, 24, 431-446.

Hay M, Polt R, Heien MH, Vanderah T, Largent-Milnes, T, Rodgers K, Victor SL, Thompson, AL, Bartlett M, Doyle K, Konhilas JP (2019) A Novel Angiotensin-(1-7)-glycosylated Mas Receptor Agonist for Treating Inflammation Related Memory Dysfunction, *Journal of Pharmacology and Experiment Therapeutics*, 369:9-25.

Hellings, A, Buchan L, Castro M, St Aubin CR, Fisher AL, Al-Nakkash L, Broderick TL and Plochocki JH. (2019). Bone strength is improved with genistein treatment in mice with diet-induced obesity. *Current Developments in Nutrition*. Oct. 23,3(11).

Henderson-Smith, A., Fisch, K.M., Hua, J., Liu, G., Ricciardelli, E., Jepsen, K., Huentelman, M., Stalberg, G., Edland, S.D., Scherzer, C.R., Dunckley, T. & Desplats, P. DNA methylation changes associated with Parkinson's disease progression: outcomes from the first longitudinal genome-wide methylation analysis in blood. *Epigenetics* 14, 365-382 (2019).

Herrera-Rivero M, Santarelli F, Brosseron F, Kummer MP, Heneka MT. Dysregulation of TLR5 and TAM Ligands in the Alzheimer's Brain as Contributors to Disease Progression. *Mol Neurobiol*. 2019 Sep;56(9):6539-6550. doi: 10.1007/s12035-019-1540-3. Epub 2019 Mar 9 (indirectly)

Hett K, Ta VT, Catheline G, Tourdias T, Manjón JV, Coupé P; Alzheimer's Disease Neuroimaging Initiative. Multimodal Hippocampal Subfield Grading For Alzheimer's Disease Classification. *Sci Rep*. 2019 Sep 25;9(1):13845. doi: 10.1038/s41598-019-49970-9. PMID: 31554909

Higbee, M, J Eliason, H Weinberg, J Lifshitz, H Handmaker. (2019) Involving Police Departments in Early Awareness of Concussion Symptoms During Domestic Violence Calls. *Journal of Aggression, Maltreatment & Trauma* 28(7): 826-837

Hodgetts CJ, Stefani M, Williams AN, Kolarik BS, Yonelinas AP, Ekstrom AD, Lawrence AD, Zhang J, and Graham KS (2019) The role of the fornix in human navigational learning. *Cortex*, 124: 97-110.

Holter M.C., Hewitt, L.T. Koebele S.V., Judd J.M., Xing L., Bimonte-Nelson H.A., Conrad C.D., Araki T., Neel B.G., Snider W.D., Newbern J.M. (2019) The Noonan Syndrome-linked Raf1L613V

mutation drives increased glial number in the mouse cortex and enhanced learning. *PLoS Genet.* 15(4). doi: 10.1371/journal.pgen.1008108. PMID: 31017896

Hou M, Grilli MD, and Glisky EL (2019) Self-reference enhances memory for multi-element events judged likely to happen in young and older adults. *Memory*, 27:1451-1461.

Huentelman M, De Both M, Jepsen W, Piras IS, Talboom JS, Willeman M, Reiman EM, Hardy J, Myers AJ. Common BACE2 Polymorphisms are Associated with Altered Risk for Alzheimer's Disease and CSF Amyloid Biomarkers in APOE  $\epsilon$ 4 Non-Carriers. *Sci Rep.* 2019 Jul 3;9(1):9640. doi: 10.1038/s41598-019-45896-4. PMID: 31270419

Huffman DJ, and Ekstrom AD (2019) A modality-independent network underlies the retrieval of large-scale spatial environments in the human brain. *Neuron*, 104:611-622.

Huffman DJ, and Ekstrom AD (2019) Which way is the bookstore? A closer look at the judgements of relative direction task. *Spatial Cognition and Computation*, 19:93-129.

Irish M, Goldberg ZL, Alaeddin S, O'Callaghan C, and Andrews-Hanna JR (2019) Age-related changes in the temporal focus and self-referential content of spontaneous cognition during periods of low cognitive demand. *Psychological Research*, 83:747-760.

Izadi A, Pevzner A, Lee DJ, Ekstrom AD, Shahlaie K, Gurkoff GG (2019) Medial Septal Stimulation Increases Seizure Threshold and Improves Cognition in Epileptic Rats. *Brain Stimulation*, 12: 735-742.

Jadavji NM, Mosnier H, Kelly E, Lawrence K, Cruickshank S, Stacey S, McCall A, Dhatt S, Arning E, Bottiglieri T, Smith PD. One-carbon metabolism supplementation improves outcome after stroke in aged male MTHFR-deficient mice. *Neurobiology of Disease*. 132: 104613.

Jadavji NM, Murray LK, Emmerson JT, Rudyk CA, Hayley S, Smith PD. A genetic deficiency in one-carbon metabolism increases oxidative stress in a mouse model of Parkinson's disease. *Toxicological Sciences*. 169:25-33. PMID: 22521626.

Jepsen, W.M., Ramsey, K., Szelinger, S., Llaci, L., Balak, C., Belnap, N., Bilagody, C., De Both, M., Gupta, R., Naymik, M., Pandey, R., Piras, I.S., Sanchez-Castillo, M., Ranganamy, S., Narayanan, V. & Huentelman, M.J. Two additional males with X-linked, syndromic mental retardation carry de novo mutations in HNRNP2. *Clin Genet* 96, 183-185 (2019).

Kaehler, BD, Bokulich, NA, McDonald D, Knight R, Caporaso, JG, Huttenhower, GA. Species abundance information improves sequence taxonomy classification accuracy. 2019. *Nature Communications*.

Kane GA, Bornstein AM, Shenhav A, Wilson RC, Daw ND, and Cohen JD (2019) Rats exhibit similar biases in foraging and intertemporal choice tasks. *eLife*, 8:e48429.

Kari E, Llaci L, Go JL, Maymik M, Knowles JA, Leal SM, Ranganamay S, Huentelman MJ, Liang W, Friedman RA, Schrauwen I (2019) Genes implicated in rare congenital inner ear and cochleovestibular nerve malformations. *Ear and Hearing* Jan 24 [online ahead of print]; PMID: 31985533.

Kari, E., Llaci, L., Go, J.L., Naymik, M., Knowles, J.A., Leal, S.M., Rangasamy, S., Huentelman, M.J., Friedman, R.A. & Schrauwen, I. A de novo SIX1 variant in a patient with a rare nonsyndromic cochleovestibular nerve abnormality, cochlear hypoplasia, and bilateral sensorineural hearing loss. *Mol Genet Genomic Med* 7, e995 (2019).

Karin R, Koc M, Rettberg JR, Hodis HN, Henderson VW, St. John JA., Allayee H., Brinton RD, Mack WJ (2019) Apolipoprotein E4 genotype in combination with poor metabolic profile is associated with reduced cognitive performance in healthy postmenopausal women: implications for late onset Alzheimer's disease. *Menopause*, 26:7-15.

Keerthivasan MB, Saranathan M., Johnson K, Fu Z, Weinkauff, CC, Martin DR, Bilgin A., and Altbach, M. I. (2019). An efficient 3D stack-of-stars turbo spin echo pulse sequence for simultaneous T2-weighted imaging and T2 mapping. *Magnetic Resonance in Medicine*, 82:326-341.

Kelley CM, Ginsberg SD, Alldred MJ, Strupp BJ, Mufson EJ. Maternal Choline Supplementation Alters Basal Forebrain Cholinergic Neuron Gene Expression in the Ts65Dn Mouse Model of Down Syndrome. *Dev Neurobiol.* 2019 Jul;79(7):664-683. doi: 10.1002/dneu.22700. Epub 2019 Jun 9. PubMed PMID: 31120189.

Kelley CM, Perez SE, Mufson EJ. Tau pathology in the medial temporal lobe of athletes with chronic traumatic encephalopathy: a chronic effect of neurotrauma consortium study. *Acta Neuropathol Commun.* 2019 Dec 12;7(1):207. PMID:31831066

Keung W, Hagen T, and Wilson RC (2019) Regulation of evidence accumulation by pupil-linked arousal processes. *Nature Human Behavior*, 3:636-645.

Kling, J.M., Dowling, N.M., Bimonte-Nelson, H.A., Gleason, C.E., Kantarci, K., Manson, J.E., Taylor, H.S., Brinton, E.A., Lobo, R.A., Cedars, M.I., Pal, L., Neal-Perry, G., Naftolin, F., Harman, S.M., Miller, V.M. (2019). Impact of menopausal hormone formulations on pituitary-ovarian regulatory feedback. *Am J Physiol Regul Integr Comp Physiol.* 317(6): R912-R920. doi: 10.1152/ajpregu.00234.2019. PMID: 31663769

Koebele, S.V., Nishimura, K.J., Bimonte-Nelson, H.A., Kemmou, S., Ortiz, J.B., Judd, J.M., Conrad, C.D. (2019). A long-term cyclic plus tonic regimen of 17 $\beta$ -estradiol improves the ability to handle a high spatial working memory load in ovariectomized middle-aged female rats. *Horm behav.* 118. doi: 10.1016/j.yhbeh.2019.104656. PMID: 31862208.

Kundu S, Lukemire J, Wang Y, Guo Y; Alzheimer's Disease Neuroimaging Initiative. A Novel Joint Brain Network Analysis Using Longitudinal Alzheimer's Disease Data. *Sci Rep.* 2019 Dec 20;9(1):19589. doi: 10.1038/s41598-019-55818-z. PMID: 31863067

Kun-Rodrigues C, Orme T, Carmona S, Hernandez DG, Ross OA, Eicher JD, Shepherd C, Parkkinen L, Darwent L, Heckman MG, Scholz SW, Troncoso JC, Pletnikova O, Dawson T, Rosenthal L, Ansorge O, Clarimon J, Lleo A, Morenas-Rodriguez E, Clark L, Honig LS, Marder K, Lemstra A, Rogava E, St George-Hyslop P, Londos E, Zetterberg H, Barber I, Braae A, Brown K, Morgan K, Troakes C, Al-Sarraj S, Lashley T, Holton J, Compta Y, Van Deerlin V, Serrano GE, Beach TG, Lesage S, Galasko D, Masliah E, Santana I, Pastor P, Diez-Fairen M, Aguilar M, Tienari PJ, Myllykangas L, Oinas M, Revesz T, Lees A, Boeve BF, Petersen RC, Ferman TJ, Escott-Price V, Graff-Radford N, Cairns NJ, Morris JC, Pickering-Brown S, Mann D, Halliday GM, Hardy J, Trojanowski JQ, Dickson DW, Singleton A, Stone DJ, Guerreiro R, Bras J. A

comprehensive screening of copy number variability in dementia with Lewy bodies. *Neurobiol Aging*. 2019 Mar;75:223.e1-223.e10. doi: 10.1016/j.neurobiolaging.2018.10.019. Epub 2018 Oct 24. PubMed PMID: 30448004.

Kyle CT, Permenter MR, Vogt JA, Rapp PR, and Barnes, C.A. (2019) Behavioral impact of long-term chronic implantation of neural recording devices in the rhesus macaque. *Neuromodulation: Technology at the Neural Interface*, 22:435-440.

Kyle CT, Stokes J, Bennett J, Meltzer J, Permenter MR, Vogt JA, Ekstrom A, and Barnes CA (2019) Cytoarchitectonically-driven MRI atlas of nonhuman primate hippocampus: preservation of subfield volumes in aging. *Hippocampus*, 29:409-421.

Langbaum JB, Karlawish J, Roberts JS, Wood EM, Bradbury A, High N, Walsh TL, Gordon D, Aggarwal R, Davis P, Stowell C, Trisko L, Langlois CM, Reiman EM, Tariot PN. GeneMatch: A novel recruitment registry using at-home APOE genotyping to enhance referrals to Alzheimer's prevention studies. *Alzheimers Dement*. 2019 Apr;15(4):515-524. doi: 10.1016/j.jalz.2018.12.007. Epub 2019 Feb 13. PMID: 30772251

Langer K, O'Shea D, De Wit L, DeFeis B, Mejia A, Amofa P, Chandler M, Locke DEC, Fields JA, Phatak V, Dean P, & Smith G. (2019). Self-efficacy mediates the association between physical function and perceived quality of life in individuals with mild cognitive impairment. *Journal of Alzheimer's Disease*, 68 (4), 1511-1519.

Langlois CM, Bradbury A, Wood EM, Roberts JS, Kim SYH, Riviere ME, Liu F, Reiman EM, Tariot PN, Karlawish J, Langbaum JB. Alzheimer's Prevention Initiative Generation Program: Development of an <i>APOE</i> genetic counseling and disclosure process in the context of clinical trials. *Alzheimers Dement (N Y)*. 2019 Nov 6;5:705-716. doi: 10.1016/j.trci.2019.09.013. eCollection 2019. PMID: 31921963

Lao Y, David J, Torosian A, Placencio V, Wang Y, Yang W, Tuli R, "Combined morphologic and metabolic pipeline for Positron emission tomography / computed tomography based radiotherapy response evaluation in locally advanced pancreatic adenocarcinoma", *Physics & Imaging in Radiation Oncology*, 2019 22:28-34

Leak RK, Frosch MP, Beach TG, Halliday GM. Alpha-synuclein: prion or prion-like? *Acta Neuropathol*. 2019 Oct;138(4):509-514. doi:10.1007/s00401-019-02057-1. Epub 2019 Aug 12. PubMed PMID: 31407028.

Lee G, Nho K, Kang B, Sohn KA, Kim D; for Alzheimer's Disease Neuroimaging Initiative. Predicting Alzheimer's disease progression using multi-modal deep learning approach. *Sci Rep*. 2019 Feb 13;9(1):1952. doi: 10.1038/s41598-018-37769-z. PMID: 30760848

Lewis, C.R., Henderson-Smith, A., Breitenstein, R.S., Sowards, H.A., Piras, I.S., Huentelman, M.J., Doane, L.D. & Lemery-Chalfant, K. Dopaminergic gene methylation is associated with cognitive performance in a childhood monozygotic twin study. *Epigenetics* 14, 310-323 (2019).

Li C, Chen L, McLeod E, Su J (2019) Dark mode plasmonic optical microcavity biochemical sensor. *Photonics Research*, 7:939-947.

Li S, Yin J, Nielsen M, Beach TG, Guo L, Shi J. Sirtuin 3 Mediates Tau Deacetylation. *J Alzheimers Dis*. 2019;69(2):355-362. doi: 10.3233/JAD-190014. PubMed PMID: 30958373.

Li Y, Yao Z, Yu Y, Fu Y, Zou Y, Hu B; Alzheimer's Disease Neuroimaging Initiative. The Influence of Cerebrospinal Fluid Abnormalities and APOE 4 on PHF-Tau Protein: Evidence From Voxel Analysis and Graph Theory. *Front Aging Neurosci.* 2019 Aug 8;11:208. doi: 10.3389/fnagi.2019.00208. eCollection 2019. PMID: 31440157

Lifshitz J, S Crabtree-Nelson, D Kozlowski. (2019) Traumatic Brain Injury in Domestic Violence. *Journal of Aggression, Maltreatment & Trauma.* 28(6): 655-659.

Lingo VanGilder J, VanGilder P. Functional differences in spatial processing along an anterior-posterior axis: are there implications for the retrosplenial cortex? *J Neurophysiol.* 2019 Nov 1;122(5):1845-1848. doi: 10.1152/jn.00385.2019. Epub 2019 Aug 7. PMID: 31389747

Liu J, Zhang B, Wilson G, Kong J; Alzheimer's Disease Neuroimaging Initiative. New Perspective for Non-invasive Brain Stimulation Site Selection in Mild Cognitive Impairment: Based on Meta- and Functional Connectivity Analyses. *Front Aging Neurosci.* 2019 Aug 27;11:228. doi: 10.3389/fnagi.2019.00228. eCollection 2019. PMID: 31551754

Llaci, L., Ramsey, K., Belnap, N., Claasen, A.M., Balak, C.D., Szelinger, S., Jepsen, W.M., Siniard, A.L., Richholt, R., Izat, T., Naymik, M., De Both, M., Piras, I.S., Craig, D.W., Huentelman, M.J., Narayanan, V., Schrauwen, I. & Rangasamy, S. Compound heterozygous mutations in SNAP29 is associated with Pelizaeus-Merzbacher-like disorder (PMLD). *Hum Genet* 138, 1409-1417 (2019).

Lopez Lopez C, Tariot PN, Caputo A, Langbaum JB, Liu F, Riviere ME, Langlois C, Rouzade-Dominguez ML, Zalesak M, Hendrix S, Thomas RG, Viglietta V, Lenz R, Ryan JM, Graf A, Reiman EM. The Alzheimer's Prevention Initiative Generation Program: Study design of two randomized controlled trials for individuals at risk for clinical onset of Alzheimer's disease. *Alzheimers Dement (N Y).* 2019 Jun 12;5:216-227. doi: 10.1016/j.trci.2019.02.005. eCollection 2019. Review. PMID: 31211217

Lorch G, Sivaprakasam K, Zismann V, Perdignes N, Murtaza M, Contente-Cuomo T, Nazareno A, Facista S, Wong S, Liang WS, Amann J, Sinicropi-yao S, Koenig M, La Perle K, Whitsett T, Trent J, Carbone D, Hendricks W (2019) Identification of frequent activating HER2 mutations in primary canine pulmonary adenocarcinoma *Clinical Cancer Res*, doi: 10.1158/1078-0432.CCR-19-1145. PMID: 31431454 [Epub ahead of print].

Lu WF, Wang L, Zhou W, and Fu W (2019) Augmented reality navigation to assist retrograde peroneal access for the endovascular treatment of critical limb ischemia. *Journal of Vascular Surgery Cases and Innovative Techniques*, 5:518-520.

Lue LF, Beach TG, Walker DG. Alzheimer's Disease Research Using Human Microglia. *Cells.* 2019 Aug 5;8(8). pii: E838. doi: 10.3390/cells8080838. Review. PubMed PMID: 31387311.

Lue LF, Kuo YM, Sabbagh M. Advance in Plasma AD Core Biomarker Development: Current Findings from Immunomagnetic Reduction-Based SQUID Technology. *Neurol Ther.* 2019 Dec;8(Suppl 2):95-111. doi: 10.1007/s40120-019-00167-2. Epub 2019 Dec 12. Review. PubMed PMID: 31833027.

Lue LF, Pai MC, Chen TF, Hu CJ, Huang LK, Lin WC, Wu CC, Jeng JS, Blennow K, Sabbagh MN, Yan SH, Wang PN, Yang SY, Hatsuta H, Morimoto S, Takeda A, Itoh Y, Liu J, Xie H, Chiu MJ. Corrigendum: Age-Dependent Relationship Between Plasma A $\beta$ 40 and A $\beta$ 42 and Total Tau

Levels in Cognitively Normal Subjects. *Front Aging Neurosci.* 2019 Nov 8;11:292. doi: 10.3389/fnagi.2019.00292. eCollection 2019. PubMed PMID: 31754353.

Lue LF, Pai MC, Chen TF, Hu CJ, Huang LK, Lin WC, Wu CC, Jeng JS, Blennow K, Sabbagh MN, Yan SH, Wang PN, Yang SY, Hatsuta H, Morimoto S, Takeda A, Itoh Y, Liu J, Xie H, Chiu MJ. Age-Dependent Relationship Between Plasma A $\beta$ 40 and A $\beta$ 42 and Total Tau Levels in Cognitively Normal Subjects. *Front Aging Neurosci.* 2019 Sep 3;11:222. doi: 10.3389/fnagi.2019.00222. eCollection 2019. Erratum in: *Front Aging Neurosci.* 2019 Nov 08;11:292. PubMed PMID: 31551751.

Lund BT, Stone R, Levy AM, Lee S, Rodgers KE, Kelland EE. (2019) Reduced disease severity following therapeutic treatment with Angiotensin 1-7 in a mouse model of multiple sclerosis, *Neurobiology of Disease*, 127:87-100, DOI: 10.1016/j.nbd.2019.02.018.

Luo M, Ma W, Sand Z, Finlayson J, Wang T, Brinton RD, Willis WT, and Mandarino LJ (2019) Von Willebrand factor A domain--containing protein 8 (VWA8) localizes to the matrix side of the inner mitochondrial membrane. *Biochem Biophys Res Commun.*, 52:158-163.

Ly CV, Koenig L, Christensen J, Gordon B, Beaumont H, Dahiya S, Chen J, Su Y, Nelson B, Jockel-Balsarotti J, Drain C, Jerome G, Morris JC, Fagan AM, Harms MB, Benzinger TLS, Miller TM, Ances BM. Tau positron emission tomography imaging in C9orf72 repeat expansion carriers. *Eur J Neurol.* 2019 Sep;26(9):1235-1239. doi: 10.1111/ene.13940. Epub 2019 Mar 25. PMID: 30790403

Mahady L, Nadeem M, Malek-Ahmadi M, Chen K, Perez SE, Mufson EJ. HDAC2 dysregulation in the nucleus basalis of Meynert during the progression of Alzheimer's disease. *Neuropathol Appl Neurobiol.* 2019 Jun;45(4):380-397. PMID:30252960.

Mak M, Lam C, Pineda SJ, Lou M, Xu LY, Meeks C, Lin C, Stone R, Rodgers K, Mitani G (2019) Pharmacogenetics of Warfarin in a Diverse Patient Population. *Journal of Cardiovascular Pharmacology and Therapeutics*, 24:521-533, doi: 10.1177/1074248419843530.

Malek-Ahmadi M, Beach TG, Zamrini E, Adler CH, Sabbagh MN, Shill HA, Jacobson SA, Belden CM, Caselli RJ, Woodruff BK, Rapsack SZ, Ahern GL, Shi J, Caviness JN, Driver-Dunckley E, Mehta SH, Shprecher DR, Spann BM, Tariot P, Davis KJ, Long KE, Nicholson LR, Intorcchia A, Glass MJ, Walker JE, Callan M, Curry J, Cutler B, Oliver J, Arce R, Walker DG, Lue LF, Serrano GE, Sue LI, Chen K, Reiman EM. Faster cognitive decline in dementia due to Alzheimer disease with clinically undiagnosed Lewy body disease. *PLoS One.* 2019; 14 (6):e0217566 Epub 2019 June 25 PMID: 31237877 PMCID: 6592515 DOI: 10.1371/journal.pone.0217566

Malek-Ahmadi M, Chen K, Perez SE, Mufson EJ. Cerebral Amyloid Angiopathy and Neuritic Plaque Pathology Correlate with Cognitive Decline in Elderly Non-Demented Individuals. *J Alzheimers Dis.* 2019;67(1):411-422. PMID:30594928.

Manivannan B, Yegambaram M, Supowit S, Beach TG, Halden RU. Assessment of Persistent, Bioaccumulative and Toxic Organic Environmental Pollutants in Liver and Adipose Tissue of Alzheimer's Disease Patients and Age-matched Controls. *Curr Alzheimer Res.* 2019;16(11):1039-1049. doi: 10.2174/1567205016666191010114744. PubMed PMID: 31660829.

Manocha GD, Floden AM, Miller NM, Smith AJ, Nagamoto-Combs K, Saito T, Saido TC, Combs CK (2019) Temporal progression of Alzheimer's disease in brains and intestines of transgenic mice. *Neurobiol Aging*. 81:166-176. (indirectly)

Manzo E, Lorenzini I, Barrameda D, O'Conner AG, Barrows JM, Starr A, Kovalik T, Rabichow BE, Lehmkuhl EM, Shreiner DD, Joardar A, Liévens JC, Bowser R, Sattler R, Zarnescu DC. Glycolysis upregulation is neuroprotective as a compensatory mechanism in ALS. *eLife* 2019 Jun 10;8. pii: e45114. doi: 10.7554/eLife.45114

Martinez, BI, Stabenfeldt, SE. Current trends in biomarker discovery and analysis tools for traumatic brain injury. *Journal of Biological Engineering*. 2019; 13: 16. DOI: 10.1186/s13036-019-0145-8

McCullough, C.G., Szelinger, S., Belnap, N., Ramsey, K., Schrauwen, I., Claasen, A.M., Burke, L.W., Siniard, A.L., Huentelman, M.J., Narayanan, V. & Craig, D.W. Utilizing RNA and outlier analysis to identify an intronic splice-altering variant in AP4S1 in a sibling pair with progressive spastic paraplegia. *Hum Mutat* (2019).

Meade CS, Bell RP, Towe SL, Chen NK, Hobkirk AL, Huettel SA (2019) Synergistic effects of marijuana abuse and HIV infection on neural activation during a cognitive interference task. *Addiction Biology*, 24:1235-1244. doi: 10.1111/adb.12678.

Mejia A, Smith G, Wicklund M, Armstrong M. Shared Decision Making in Mild Cognitive Impairment. *Neurology: Clinical Practice* 2019 April; 9(2): 160-164.

Memel M, Wank AA, Ryan L, Grilli MD (2019) The relationship between episodic detail generation and anterotemporal, posteromedial, and hippocampal white matter tracts. *Cortex*, 1223:124-140.

Memel M, Woolverton CB, Bourassa K, and Glisky EL (2019) Working memory predicts subsequent episodic memory decline during healthy cognitive aging: evidence from a cross-lagged panel design. *Aging, Neuropsychology and Cognition*, 26:711-730.

Mendsaikhan A, Tooyama I, Bellier JP, Serrano GE, Sue LI, Lue LF, Beach TG, Walker DG. Characterization of lysosomal proteins Progranulin and Prosaposin and their interactions in Alzheimer's disease and aged brains: increased levels correlate with neuropathology. *Acta Neuropathol Commun*. 2019 Dec 21;7(1):215. doi: 10.1186/s40478-019-0862-8. PubMed PMID: 31864418.

Miller, SJ, Philips T, Kim N, Dastgheyb R, Chen Z, Hsieh YC, Daigle JG, Datta M, Chew J, Vidensky S, Pham JT, Hughes EG, Robinson MB, Sattler R, Tomer R, Soo Suk J, Bergles DW, Haughey N, Pletnikov M, Hanes J, Rothstein JD. Molecularly defined cortical astroglia subpopulation modulates neurons via secretion of Norrin. *Nature Neuroscience*, 2019 May;22(5):741-752. doi: 10.1038/s41593-019-0366-7. Epub 2019 Apr 1

Moftah M, Emmerson JT, Jadavji NM. Linking homocysteine, B-vitamins, and choline to ischemic stroke risk. *Molecular Nutrition Vitamins*. Editor: Vinood Patel. Elsevier.

Moftah M, Jadavji NM. The role of behavioral training in reducing functional impairments after stroke. *Neural Regeneration Research*. 14:1507-08. PMID: 31089041.

Moore S, Alsop E, Lorenzini I, Starr A, Rabichow BE, Mendez E, Levy JL, Burciu C, Reiman R, Chew J, Belzil VV, Dickson DW, Robertson J, Staats KA, Ichida JK, Petrucelli L, Van Keuren-Jensen K, Sattler R. ADAR2 mislocalization and widespread RNA editing aberrations in C9orf72-mediated ALS/FTD. *Acta Neuropathologica* 2019 Jul;138(1):49-65. doi: 10.1007/s00401-019-01999-w. Epub 2019 Apr 3

Moran C, Beare R, Wang W, Callisaya M, Srikanth V; Alzheimer's Disease Neuroimaging Initiative (ADNI). Type 2 diabetes mellitus, brain atrophy, and cognitive decline. *Neurology*. 2019 Feb 19;92(8):e823-e830. Epub 2019 Jan 23. PMID: 30674592

Mueller S, Jain P, Liang WS, Kilburn L, Kline C, Gupta N, Panditharatna E, Magge SN, Zhang B, Zhu Y, Crawford JR, Banerjee A, Nazemi K, Packer RJ, Petritsch CK, Truffaux N, Roos A, Nasser S, Phillips JJ, Solomon D, Molinaro A, Waanders AJ, Byron SA, Berens ME, Kuhn J, Nazarian J, Prados M, Resnick AC. (2019) A pilot precision medicine trial for children with diffuse intrinsic pontine glioma – PNO003: a report from the Pacific Pediatric Neuro-Oncology Consortium. *Int J Cancer* Mar 12 [epub ahead of print]; PMID: 30861105.

Mufson EJ, Counts SE, Ginsberg SD, Mahady L, Perez SE, Massa SM, Longo FM, Ikonovic MD. Nerve Growth Factor Pathobiology During the Progression of Alzheimer's Disease. *Front Neurosci*. 2019 Jul 1;13:533. doi:10.3389/fnins.2019.00533. eCollection 2019. Review. PubMed PMID: 31312116.

Munger, E.L., Edler, M.K., Hopkins, W.D., Ely, J.J, Erwin, J.M., Perl, D.P., Mufson EJ, Hof PR, Sherwood, C.C. and M.A. Raghanti Astrocytic changes with aging and Alzheimer's disease-type pathology in chimpanzees. *J Comp Neurol*. 527:1179-1195, 2019 doi: 10.1002/cne.24610. PMID: 30578640.

Murray LK, Jadavji NM. (2019). The role of B-vitamin metabolism in Parkinson's disease onset, pathology, and mechanisms. *Nutrition Research Reviews*. 32: 218-230. PMID: 31303188.

Namba, M.D., Kupchik, Y.M., Spencer, S.M., Garcia-Keller, C., Goenaga, J.G., Powell, J.L., Vicino, I.A., Hogue, I.B., Gipson, C.D. (2019). Accumbens Neuroimmune Signaling and Dysregulation of Astrocytic Glutamate Transport Underlie Conditioned Nicotine Seeking Behavior. *Addiction Biology*. e12797. doi: 10.1111/adb.1279.

Narang Pp, Chen M, Sharma AA, Anderson KS and Wilson MA. 2019. The neoepitope landscape of breast cancer: implications for immunotherapy. *BMC Cancer* 19(1):200.

Nathan E. Stone, Amalee E. Nunnally, Victor Jimenez Jr, Emily K. Cope, Jason W. Sahl, Krystal Sheridan, Heidie M. Hornstra, Jacob Vinocurd, Erik W. Settles, Kyle C. Headley, Charles H.D. Williamson, Jai Ram Rideout, Evan Bolyen, J. Gregory Caporaso, Joel Terriquez, Fernando P. Monroy, Joseph D. Busch, Paul Keim, David M. Wagner. 2019. Domestic Canines do not Display Evidence of Gut Microbial Dysbiosis in the Presence of *Clostridioides* (*Clostridium*) *difficile*, Despite Cellular Susceptibility to its Toxins. *Anaerobe*. 58(53-72).

Natri Hp, Garcia A, Buetow KH, Trumble B and Wilson MA. 2019. Endogenous retroviruses and the Pregnancy Compensation Hypothesis. *Trends in Genetics* 36(1):2-3. doi: 10.1016/j.tig.2019.10.006

Natri Hp, Garcia A, Buetow KH, Trumble B and Wilson MA. 2019. The Pregnancy Pickle: Evolved immune compensation due to pregnancy underlies sex differences in human diseases. *Trends in Genetics* 35(7):478-488.

Natri Hp, Wilson MA\*, and Buetow K. 2019. Distinct molecular etiologies of male and female hepatocellular carcinoma. *BMC Cancer*. 19(951). doi: 10.1186/s12885-019-6167-2

Nguyen P-D, Zhang X, Su J (2019) One-step controlled synthesis of size-tunable toroidal gold particles for biochemical sensing, *ACS Applied Nano Materials*, 2:7839-7847.

Nielsen C, Ratiu I, Esfandiarei M, Chen A, Tierney ESS. (2019). A review of psychosocial factors of Marfan syndrome: Adolescents, adults, family and providers. *Journal of Pediatric Genetics*: 8(3): 109-122.

Nori VS, Hane CA, Crown WH, Au R, Burke WJ, Sanghavi DM, Bleicher P. Machine learning models to predict onset of dementia: A label learning approach. *Alzheimers Dement (N Y)*. 2019 Dec 10;5:918-925. doi: 10.1016/j.trci.2019.10.006. eCollection 2019. PMID: 31879701

O'Callaghan C, Shine M, Andrews-Hanna JR, and Irish M (2019) Hippocampal atrophy and intrinsic brain network dysfunction relate to alterations in mind wandering in neurodegeneration. *PNAS*, 116: 3316-3321.

Ozgur E, Roberts KE, Ozgur EO, Gin AN, Bankhead JR, Wang Z, Su J (2019) Ultra-sensitive detection of human chorionic gonadotropin using frequency locked microtoroid optical resonators. *Analytical Chemistry*, 91:11872-11878.

Palade, J, Pal, A, Rawls, A, Stabenfeldt, SE, Wilson-Rawls, J. Molecular analysis of muscle progenitor cells on extracellular matrix coatings and hydrogels. *Acta Biomaterialia*. 2019; 97: 296-309. doi: 10.1016/j.actbio.2019.08.019.

Pandit V, Lee A, Zeeshan M, Goshima K, Tan TW, Jhaji S, Trinidad B, Weinkauff C, and Zhou W (2019) The impact of frailty syndrome on the outcomes of patients with carotid stenosis, *Journal of Vascular Surgery*, 2019(19); PMID:31668557

Perez SE, Miguel JC, He B, Malek-Ahmadi M, Abrahamson EE, Ikonovic MD, Lott I, Doran E, Alldred MJ, Ginsberg SD, Mufson EJ. Frontal cortex and striatal cellular and molecular pathology in individuals with Down syndrome with and without dementia. *Acta Neuropathol*. 2019 Mar;137(3):413-436. PMID:30734106

Peter, B., Dinu, V., Liu, L., Huentelman, M., Naymik, M., Lancaster, H., Vose, C. & Schrauwen, I. Exome Sequencing of Two Siblings with Sporadic Autism Spectrum Disorder and Severe Speech Sound Disorder Suggests Pleiotropic and Complex Effects. *Behav Genet* 49, 399-414 (2019).

Piras IS, Krate J, Delvaux E, Nolz J, De Both MD, Mastroeni DF, Serrano GE, Sue LI, Beach TG, Coleman PD, Huentelman MJ. Association of AEBP1 and NRN1 RNA expression with Alzheimer's disease and neurofibrillary tangle density in middle temporal gyrus. *Brain Res*. 2019 Sep 15;1719:217-224. doi:10.1016/j.brainres.2019.06.004. Epub 2019 Jun 6. PubMed PMID: 31176712.

Piras, I.S., Krate, J., Delvaux, E., Nolz, J., Mastroeni, D.F., Persico, A.M., Jepsen, W.M., Beach, T.G., Huentelman, M.J. & Coleman, P.D. Transcriptome Changes in the Alzheimer's Disease

Middle Temporal Gyrus: Importance of RNA Metabolism and Mitochondria-Associated Membrane Genes. *J Alzheimers Dis* 70, 691-713 (2019).

Planche V, Su JH, Mournet S, Saranathan M, Dousset V, Han M, Rutt BK, Tourdias T. White-matter-nulled MPRAGE at 7T reveals thalamic lesions and atrophy of specific thalamic nuclei in multiple sclerosis. *Mult Scler*. 2019 Feb 7:1352458519828297. doi: 10.1177/1352458519828297. [Epub ahead of print] PubMed PMID: 30730233; PubMed Central PMCID: PMC6685761.

Pottier C, Ren Y, Perkerson RB 3rd, Baker M, Jenkins GD, van Blitterswijk M, DeJesus-Hernandez M, van Rooij JGJ, Murray ME, Christopher E, McDonnell SK, Fogarty Z, Batzler A, Tian S, Vicente CT, Matchett B, Karydas AM, Hsiung GR, Seelaar H, Mol MO, Finger EC, Graff C, Oijerstedt L, Neumann M, Heutink P, Synofzik M, Wilke C, Prudlo J, Rizzu P, Simon-Sanchez J, Edbauer D, Roeber S, Diehl-Schmid J, Evers BM, King A, Mesulam MM, Weintraub S, Geula C, Bieniek KF, Petrucelli L, Ahern GL, Reiman EM, Woodruff BK, Caselli RJ, Huey ED, Farlow MR, Grafman J, Mead S, Grinberg LT, Spina S, Grossman M, Irwin DJ, Lee EB, Suh E, Snowden J, Mann D, Ertekin-Taner N, Uitti RJ, Wszolek ZK, Josephs KA, Parisi JE, Knopman DS, Petersen RC, Hodges JR, Piguet O, Geier EG, Yokoyama JS, Rissman RA, Rogaeva E, Keith J, Zinman L, Tartaglia MC, Cairns NJ, Cruchaga C, Ghetti B, Kofler J, Lopez OL, Beach TG, Arzberger T, Herms J, Honig LS, Vonsattel JP, Halliday GM, Kwok JB, White CL 3rd, Gearing M, Glass J, Rollinson S, Pickering-Brown S, Rohrer JD, Trojanowski JQ, Van Deerlin V, Bigio EH, Troakes C, Al-Sarraj S, Asmann Y, Miller BL, Graff-Radford NR, Boeve BF, Seeley WW, Mackenzie IRA, van Swieten JC, Dickson DW, Biernacka JM, Rademakers R. Genome-wide analyses as part of the international FTLT-DTP whole-genome sequencing consortium reveals novel disease risk factors and increases support for immune dysfunction in FTLT. *Acta Neuropathol*. 2019 Jun; 137(6):879-899 Epub 2019 Feb 09 PMID: 30739198 PMCID: 6533145 DOI: 10.1007/s00401-019-01962-9

Prieur E, Jadavji NM\*. Assessing spatial working memory using the spontaneous y-maze alternation test in mice. *Bio Protocols*, 9:1-10.

Pyon W, Gray DT, and Barnes CA (2019) An alternative to dye-based approaches to remove background autofluorescence from primate brain tissue. *Frontiers In Neuroanatomy*, 13:73, doi: 10.3389/fnana.2019.00073.

Quarles CC, Bell LC, Stokes AM. "Imaging vascular and hemodynamic features of the brain using dynamic susceptibility contrast and dynamic contrast enhanced MRI" *Neuroimage* 187:32-55 (2019).

Rahman A, Jackson H, Hristov H, Isaacson R, Saif N, Shetty T, Etingin O, Henchcliffe C, Brinton RD, Mosconi L (2019) Sex and gender driven modifiers of Alzheimer's: The role for estrogenic control across age, race, medical, and lifestyle risks. *Frontiers in Aging Neuroscience*, doi: 10.3389/fnagi.2019.00315.

Raichlen DA, Klimentidis YC, Bharadwaj PK, Alexander GE. Differential associations of engagement in physical activity and estimated cardiorespiratory fitness with brain volume in middle-aged to older adults. *Brain Imaging Behav*. 2019 Jun 17. doi: 10.1007/s11682-019-00148-x. [Epub ahead of print] PubMed PMID: 31209836; PubMed Central PMCID: PMC7008962.

Raichlen DA, Klimentidis YC, Hsu C-H, and Alexander GE. (2019) Fractal complexity of daily physical activity patterns differs with age over the lifespan and is associated with mortality in older

adults. *Journal of Gerontology, Series A: Biological Sciences and Medical Sciences*. of *Gerontology, Series A: Biological Sciences and Medical Sciences*, 74:1461-1467.

Ralston, B, J Rable, H Handmaker, T Larson, J Lifshitz. (2019) Forensic Nursing Examination to Screen for Traumatic Brain Injury Following Intimate Partner Violence. *Journal of Aggression, Maltreatment & Trauma* 28(6): 732-743.

Rapcsak SZ (2019) Face recognition. *Current Neurology and Neuroscience Reports*, 19:41, <https://doi.org/10.1007/s11910-019-0960-9>.

Readhead B, Haure-Mirande JV, Ehrlich ME, Gandy S, Dudley JT. 2019a. Clarifying the Potential Role of Microbes in Alzheimer's Disease. *Neuron*, 104, 1036-1037.

Readhead B, Haure-Mirande JV, Ehrlich ME, Gandy S, Dudley JT. 2019b. Further evidence of increased human Herpesvirus in Alzheimer's disease. *BioRxiv*, 858050.

Reddy R, Liu S, Asante I, Parikh P, Rodgers K, Liebler J, Borok A, Baydur A, Louie S (2019) Circulating Angiotensin Peptides Levels in Acute Respiratory Distress Syndrome Correlate with Clinical Outcomes: A Pilot Study. *PLOS*, 2019 Mar 7;14(3):e0213096. doi: 10.1371/journal.pone.0213096. eCollection 2019. PMID: 30845246

Reiman EM. Editorial: Blood Tests for Alzheimer's Disease and Related Disorders. *J Prev Alzheimers Dis*. 2019;6(3):155-156. doi: 10.14283/jpad.2019.20. No abstract available. PMID: 31062824

Revill AL, Chu NY, Ma L, LeBlancq MJ, Dickson CT, Funk GD. (2019). Postnatal development of persistent inward currents in rat XII motoneurons and their modulation by serotonin, muscarine and norepinephrine. *Journal of Physiology*. 597(12): 3183-3201.

Rockwood S, Mason D, Lord R, Lamar P, Prozialeck W, Banayat T and Al-Nakkash L. (2019). Genistein diet improves body weight, serum glucose and triglyceride levels in both male and female ob/ob mice. *Diabetes Metabolic Syndrome & Obesity*. 12: 2011-2021.

Rowe, RK, JL Harrison, HW Morrison, V Subbian, SM Murphy, J Lifshitz. (2019) Acute Post-Traumatic Sleep May Define Vulnerability to a Second Traumatic Brain Injury in Mice. *Journal of Neurotrauma* 36(8): 1318-1334. PMID: 30398389

Ryan, L., Hay, M., Huentelman, M.J., Duarte, A., Rundek, T., Levin, B., Soldan, A., Pettigrew, C., Mehl, M.R. & Barnes, C.A. Precision Aging: Applying Precision Medicine to the Field of Cognitive Aging. *Front Aging Neurosci* 11, 128 (2019).

Sabat J, Bock D, Hsu C H, Tan TW, Weinkauff C., Trouard T, Perez-Carrillo GG, and Zhou W. (2019) Risk factors associated with microembolization after carotid intervention. *Journal of Vascular Surgery*, doi: 10.1016/j.jvs.2019.06.202. [Epub ahead of print].

Sabat J, Hsu C, Samra N, Chu, Q, Weinkauff C, Goshima G, Zhou W, and Tan TW (2019) Length of stay and ICU stay are increase with repair of traumatic superior mesenteric vein Injury. *Journal of Surgical Research*, 242: 94-99

Saber M, Giordano KR, Hur Y, Ortiz JB, Morrison H, Godbout JP, Murphy SM, Lifshitz J, Rowe RK. Acute peripheral inflammation and post-traumatic sleep differ between sexes after

experimental diffuse brain injury. *Eur J Neurosci.* 2019 Nov 2. doi: 10.1111/ejn.14611. [Epub ahead of print] PubMed PMID: 31677290

Sabir MS, Blauwendraat C, Ahmed S, Serrano GE, Beach TG, Perkins M, Rice AC, Masliah E, Morris CM, Pihlstrom L, Pantelyat A, Resnick SM, Cookson MR, Hernandez DG, Albert M, Dawson TM, Rosenthal LS, Houlden H, Pletnikova O, Troncoso J, Scholz SW. Assessment of APOE in atypical parkinsonism syndromes. *Neurobiol Dis.* 2019 Jul;127:142-146. doi: 10.1016/j.nbd.2019.02.016. Epub 2019 Feb 21. PubMed PMID: 30798004.

Sales E and Chou Y-H (2019) Yoga as advanced cognitive training: How Kirtan Kriya in Kundalini Yoga changes the PTSD brain. *Journal of Yoga and Physiotherapy*, 7(4): JYP.MS.ID.555721.

Satizabal, C.L., Adams, H.H.H., Hibar, D.P., White, C.C., Knol, M.J., Stein, J.L., Scholz, M., Sargurupremraj, M., Jahanshad, N., Roshchupkin, G.V., Smith, A.V., Bis, J.C., Jian, X., Luciano, M., Hofer, E., Teumer, A., van der Lee, S.J., Yang, J., Yanek, L.R., Lee, T.V., Li, S., Hu, Y., Koh, J.Y., Eicher, J.D., Desrivieres, S., Arias-Vasquez, A., Chauhan, G., Athanasiu, L., Renteria, M.E., Kim, S., Hoehn, D., Armstrong, N.J., Chen, Q., Holmes, A.J., den Braber, A., Kloszewska, I., Andersson, M., Espeseth, T., Grimm, O., Abramovic, L., Alhusaini, S., Milaneschi, Y., Pampmeyer, M., Axelsson, T., Ehrlich, S., Roiz-Santianez, R., Kraemer, B., Haberg, A.K., Jones, H.J., Pike, G.B., Stein, D.J., Stevens, A., Bralten, J., Vernooij, M.W., Harris, T.B., Filippi, I., Witte, A.V., Guadalupe, T., Wittfeld, K., Mosley, T.H., Becker, J.T., Doan, N.T., Hagenaars, S.P., Saba, Y., Cuellar-Partida, G., Amin, N., Hilal, S., Nho, K., Mirza-Schreiber, N., Arfanakis, K., Becker, D.M., Ames, D., Goldman, A.L., Lee, P.H., Boomsma, D.I., Lovestone, S., Giddaluru, S., Le Hellard, S., Mattheisen, M., Bohlken, M.M., Kasperaviciute, D., Schmaal, L., Lawrie, S.M., Agartz, I., Walton, E., Tordesillas-Gutierrez, D., Davies, G.E., Shin, J., Ipser, J.C., Vinke, L.N., Hoogman, M., Jia, T., Burkhardt, R., Klein, M., Crivello, F., Janowitz, D., Carmichael, O., Haukvik, U.K., Aribisala, B.S., Schmidt, H., Strike, L.T., Cheng, C.Y., Risacher, S.L., Putz, B., Fleischman, D.A., Assareh, A.A., Mattay, V.S., Buckner, R.L., Mecocci, P., Dale, A.M., Cichon, S., Boks, M.P., Matarin, M., Penninx, B., Calhoun, V.D., Chakravarty, M.M., Marquand, A.F., Macare, C., Kharabian Masouleh, S., Oosterlaan, J., Amouyel, P., Hegenscheid, K., Rotter, J.I., Schork, A.J., Liewald, D.C.M., de Zubicaray, G.I., Wong, T.Y., Shen, L., Samann, P.G., Brodaty, H., Roffman, J.L., de Geus, E.J.C., Tsolaki, M., Erk, S., van Eijk, K.R., Cavalleri, G.L., van der Wee, N.J.A., McIntosh, A.M., Gollub, R.L., Bulayeva, K.B., Bernard, M., Richards, J.S., Himali, J.J., Loeffler, M., Rommelse, N., Hoffmann, W., Westlye, L.T., Valdes Hernandez, M.C., Hansell, N.K., van Erp, T.G.M., Wolf, C., Kwok, J.B.J., Vellas, B., Heinz, A., Olde Loohuis, L.M., Delanty, N., Ho, B.C., Ching, C.R.K., Shumskaya, E., Singh, B., Hofman, A., van der Meer, D., Homuth, G., Psaty, B.M., Bastin, M.E., Montgomery, G.W., Foroud, T.M., Reppermund, S., Hottenga, J.J., Simmons, A., Meyer-Lindenberg, A., Cahn, W., Whelan, C.D., van Donkelaar, M.M.J., Yang, Q., Hosten, N., Green, R.C., Thalamuthu, A., Mohnke, S., Hulshoff Pol, H.E., Lin, H., Jack, C.R., Jr., Schofield, P.R., Muhleisen, T.W., Maillard, P., Potkin, S.G., Wen, W., Fletcher, E., Toga, A.W., Gruber, O., Huentelman, M., Davey Smith, G., Launer, L.J., Nyberg, L., Jonsson, E.G., Crespo-Facorro, B., Koen, N., Greve, D.N., Uitterlinden, A.G., Weinberger, D.R., Steen, V.M., Fedko, I.O., Groenewold, N.A., Niessen, W.J., Toro, R., Tzourio, C., Longstreth, W.T., Jr., Ikram, M.K., Smoller, J.W., van Tol, M.J., Sussmann, J.E., Paus, T., Lemaitre, H., Schroeter, M.L., Mazoyer, B., Andreassen, O.A., Holsboer, F., Depondt, C., Veltman, D.J., Turner, J.A., Pausova, Z., Schumann, G., van Rooij, D., Djurovic, S., Deary, I.J., McMahon, K.L., Muller-Myhsok, B., Brouwer, R.M., Soininen, H., Pandolfo, M., Wassink, T.H., Cheung, J.W., Wolfers, T., Martinot, J.L., Zwiers, M.P., Nauck, M., Melle, I., Martin, N.G., Kanai, R., Westman, E., Kahn, R.S., Sisodiya, S.M., White, T., Saremi, A., van Bokhoven, H., Brunner, H.G., Volzke, H., Wright, M.J., van 't Ent, D., Nothen, M.M., Ophoff, R.A., Buitelaar, J.K., Fernandez, G., Sachdev, P.S., Rietschel, M., van Haren, N.E.M., Fisher, S.E., Beiser, A.S., Francks, C., Saykin, A.J., Mather,

K.A., Romanczuk-Seiferth, N., Hartman, C.A., DeStefano, A.L., Heslenfeld, D.J., Weiner, M.W., Walter, H., Hoekstra, P.J., Nyquist, P.A., Franke, B., Bennett, D.A., Grabe, H.J., Johnson, A.D., Chen, C., van Duijn, C.M., Lopez, O.L., Fornage, M., Wardlaw, J.M., Schmidt, R., DeCarli, C., De Jager, P.L., Villringer, A., Debette, S., Gudnason, V., Medland, S.E., Shulman, J.M., Thompson, P.M., Seshadri, S. & Ikram, M.A. Genetic architecture of subcortical brain structures in 38,851 individuals. *Nat Genet* 51, 1624-1636 (2019).

Savica R, Beach TG, Hentz JG, Sabbagh MN, Serrano GE, Sue LI, Dugger BN, Shill HA, Driver-Dunckley E, Caviness JN, Mehta SH, Jacobson SA, Belden CM, Davis KJ, Zamrini E, Shprecher DR, Adler CH. Lewy body pathology in Alzheimer's disease: A clinicopathological prospective study. *Acta Neurol Scand*. 2019 Jan;139(1):76-81. doi: 10.1111/ane.13028. Epub 2018 Oct 7. PubMed PMID: 30229861.

Schedlbauer AM and Ekstrom AD (2019) Flexible network community organization during encoding and retrieval of spatiotemporal episodic memories. *Network Neuroscience*, 3:1070-1093.

Schminda KM, Prah MA, Hu LS, Quarles CC, Semmineh NB, Rand SD, Connelly JM, Anderies B, Zhou Y, Liu Y, Logan B, Stokes AM, Baird G, Boxerman JL. "Moving Toward a Consensus DSC-MRI Protocol: Validation of a Low-Flip Angle Single-Dose Option as a Reference Standard for Brain Tumors" *AJNR* 626-633 (2019).

Schneider LS, Hernandez G, Zhao L, Franke AA, Chen YL, Pawluczyk S, Mack WJ, Brinton RD (2019) Safety and feasibility of estrogen receptor-- $\beta$  targeted phytoSERM formulation for menopausal symptoms: phase 1b/2a randomized clinical trial. *Menopause*, 26:874-884.

Schrauwen, I., Valgaeren, H., Tomas-Roca, L., Sommen, M., Altunoglu, U., Wesdorp, M., Beyens, M., Fransen, E., Nasir, A., Vandeweyer, G., Schepers, A., Rahmoun, M., van Beusekom, E., Huentelman, M.J., Offeciers, E., Dhooghe, I., Huber, A., Van de Heyning, P., Zanetti, D., De Leenheer, E.M.R., Gilissen, C., Hoischen, A., Cremers, C.W., Verbist, B., de Brouwer, A.P.M., Padberg, G.W., Pennings, R., Kayserili, H., Kremer, H., Van Camp, G. & van Bokhoven, H. Variants affecting diverse domains of MEPE are associated with two distinct bone disorders, a craniofacial bone defect and otosclerosis. *Genet Med* 21, 1199-1208 (2019).

Schultz SA, Gordon BA, Mishra S, Su Y, Morris JC, Ances BM, Duchek JM, Balota DA, Benzinger TLS. Association between personality and tau-PET binding in cognitively normal older adults. *Brain Imaging Behav*. 2019 Sep 5. doi: 10.1007/s11682-019-00163-y. [Epub ahead of print] PMID: 31486975

Scott DD, Francois-Moutal L, Kumirov VK, Khanna M (2019) 1H, 15N and 13C backbone assignment of apo TDP-43 RNA recognition motifs. *Biomolecular NMR Assignments*, 13:163-167.

Sekar S, Geiger P, Cuyugan L, Boyle A, Serrano G, Beach TG, Liang WS. Identification of Circular RNAs using RNA Sequencing. *J Vis Exp*. 2019 Nov 14;(153). doi: 10.3791/59981. PubMed PMID: 31789321.

Seo K, Pan R, Lee D, Thiyyagura P, Chen K; Alzheimer's Disease Neuroimaging Initiative. Visualizing Alzheimer's disease progression in low dimensional manifolds. *Heliyon*. 2019 Aug 2;5(8):e02216. doi: 10.1016/j.heliyon.2019.e02216. eCollection 2019 Aug. PMID: 31406946

Shaughnessy L, Sheard S, Goldfarb D, Atri A. Cognitive Assessment of Alzheimer's Disease and Dementias in Clinical Practice: Pragmatics of Brief Instruments and Neuropsychological Evaluation. *J Clin Psychiatry*. 2019 Jun 25;80(4). pii: MS18002BR2C. doi: 10.4088/JCP.MS18002BR2C. PubMed PMID: 31237995.

Shoeibi A, Litvan I, Juncos JL, Bordelon Y, Riley D, Standaert D, Reich SG, Shprecher D, Hall D, Marras C, Kluger B, Olfati N, Jankovic J. Are the International Parkinson disease and Movement Disorder Society progressive supranuclear palsy (IPMDS-PSP) diagnostic criteria accurate enough to differentiate common PSP phenotypes? *Parkinsonism Relat Disord*. 2019 Dec;69:34-39. doi: 10.1016/j.parkreldis.2019.10.012. Epub 2019 Oct 14. PubMed PMID: 31665686.

Shprecher D, Zhang N, Halverson M, Savica R (2019). Parkinsonism Risk Factors in Salt Lake City, Utah: A Community-Based Study. *Brain Sci*, 9(3).

Siemsen, B.M., Reichel, C.M., Leong, J.C., Garcia-Keller, C., Gipson, C.D., Spencer, S.M., McFaddin, J.A., Hooker, K.N., Kalivas, P.W., Scofield, M.D. (2019). Effects of methamphetamine self-administration and extinction on astrocyte structure and function in the nucleus accumbens core. *Neuroscience*, doi: 10.1016/j.neuroscience.2019.03.040. PMID: 30926546.

Smith R, Ahern GL, and Lane RD. (2019) The role of the anterior and midcingulate cortex in emotional awareness: A domain-general processing perspective. In: *Handbook of Clinical Neurology*, Vol 166, 3rd Series, B Vogt (Ed): Cingulate Cortex. Cambridge, Elsevier, pg. 89-101.

Soman S, Dai W, Dong L, Hitchner E, Lee K, Baughman B, Holdsworth S, Massaband P, Bhat J, Mosely M, Rosen A, Zhou W, and Zaharchuk G (2019) Identifying cardiovascular risk factors that impact cerebrovascular reactivity: an ASL MRI study. *Journal of Magnetic Resonance Imaging*, <https://doi.org/10.1002/jmri.26862>.

Soto M, Gaffney KJ, Rodgers KE. (2019) Improving the innate immune response in diabetes by Modifying the renin angiotensin system. *Frontiers in Immunology*, 10:2885, doi: 10.3389/fimmu.2019.02885. eCollection.

Soto S, Gaffney KJ, and Rodgers KE (2019) Immunosuppression in T2D is Improved with RAS – modifying treatment in a *S. aureus* infection model. *Frontiers in Immunology*, doi: 10.3389/fimmu.2019.02885.

Sripasert I, Mack WJ, Hodis HN, Allayee H, Brinton RD, and Karim R (2019) Effect of ApoE4 genotype on the association between metabolic phenotype and subclinical atherosclerosis in postmenopausal women. *American Journal of Cardiology*, 124:1031-1037.

Standage-Beier K, Brookhouser N, Balachandran P, Zhang Q, Brafman DA, Wang X. RNA-Guided Recombinase-Cas9 fusion targets genomic DNA deletion and integration. *CRISPR J*. 2019 Aug;2:209-222.

Starrett MJ, Stokes JD, Huffman DJ, Ferrer E, Ekstrom AD (2019) Learning-Dependent Evolution of Spatial Representations in Large-Scale Virtual Environments. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, 45:497-514

Stelzer E-M, Knowles, LM, Wilson DT, O'Connor, MF (2019) Recruitment and retention in clinical and experimental bereavement research: Lessons learned from creating a research registry. *Death Studies*, DOI 10.1080/07481187.2019.160913.

Stern RA, Adler CH, Chen K, Navitsky M, Luo J, Dodick DW, Alosco ML, Tripodis Y, Goradia DD, Martin B, Mastroeni D, Fritts NG, Jarnagin J, Devous MD Sr, Mintun MA, Pontecorvo MJ, Shenton ME, Reiman EM. Tau Positron-Emission Tomography in Former National Football League Players. *N Engl J Med*. 2019 May 2;380(18):1716-1725. doi: 10.1056/NEJMoa1900757. Epub 2019 Apr 10. PMID: 30969506

Stern Y, Barnes CA, Grady C, Jones RN, Raz N (2019) Brain reserve, cognitive reserve, compensation, and maintenance: operationalization, validity, and mechanisms of cognitive resilience. *Neurobiology of Aging*, 83:124-129.

Stewart, W, ... J Lifshitz, ... DH Smith. (2019) Primum Non Nocere: A Call for Balance when Reporting on Chronic Traumatic Encephalopathy. *Lancet Neurology* 18(3): 231-233.PMID: 30784550

Stickel A, McKinnon A, Ruiz J, Grilli MD, and Ryan L (2019) The impact of cardiovascular risk factors on cognition in Hispanics and non-Hispanic Whites. *Learning & Memory*, 26:235-244

Stone RE, Liu S, Levy AM, Kashani N, Louie S, Rodgers KE, Kelland EE, Lund BT. (2019) Activation of the protective arm of the renin angiotensin system in demyelinating disease. *Neurobiology of Disease, Journal of Neuroimmune Pharmacology*, doi.org/10.1007/s11481-019-09894-7.

Stonnington CM, Krell-Roesch J, Locke DEC, Hentz JG, Dueck AC, Geda YE, Tariot PN, Caselli RJ. Impact of Zumba on Cognition and Quality of Life is Independent of APOE4 Carrier Status in Cognitively Unimpaired Older Women: A 6-Month Randomized Controlled Pilot Study. *Am J Alzheimers Dis Other Dement*. 2019 Aug 11; 1533317519868370 [Epub ahead of print] PMID: 31401841 DOI: 10.1177/1533317519868370

Strikwerda-Brown C, Grilli MD, Andrews-Hanna J, and Irish M (2019) 'All is not lost' – Rethinking the nature of memory and the self in dementia. *Ageing Research Reviews*, 54:100932.

Su J, Thomas FT, Kasoff WS, Tourdias T, Choi EY, Rutt BK, and Saranathan M. (2019) Thalamus optimized multi-atlas segmentation (THOMAS): fast, fully automated segmentation of thalamic nuclei from anatomical MRI. *Neuroimage*. 194:272-282.

Su Y, Flores S, Wang G, Hornbeck RC, Speidel B, Joseph-Mathurin N, Vlassenko AG, Gordon BA, Koeppe RA, Klunk WE, Jack CR Jr, Farlow MR, Salloway S, Snider BJ, Berman SB, Roberson ED, Brosch J, Jimenez-Velazques I, van Dyck CH, Galasko D, Yuan SH, Jayadev S, Honig LS, Gauthier S, Hsiung GR, Masellis M, Brooks WS, Fulham M, Clarnette R, Masters CL, Wallon D, Hannequin D, Dubois B, Pariente J, Sanchez-Valle R, Mummery C, Ringman JM, Bottlaender M, Klein G, Milosavljevic-Ristic S, McDade E, Xiong C, Morris JC, Bateman RJ, Benzinger TLS. Comparison of Pittsburgh compound B and florbetapir in cross-sectional and longitudinal studies. *Alzheimers Dement (Amst)*. 2019 Feb 22;11:180-190. doi: 10.1016/j.dadm.2018.12.008. eCollection 2019 Dec. PMID: 30847382

Sukhina, AS, SD Brown, KS Lewis, OJ Oatman, TC Thomas, RK Rowe, PD Adelson, J Lifshitz. (in press) Failure to Thrive in a 15 Month-Old with a History of Head Trauma. *Pediatrics in Review*. (PIR-2018-0069)

Talboom JS, Håberg AK, De Both MD, Naymik MA, Schrauwen I, Lewis DR, Bertinelli SF, Hammersland C, Fritz MA, Myers A, Hay M, Barnes CA, Glisky E, Ryan L, and Huentelman MJ

(2019) Family history of Alzheimer's disease alters cognition and is modified by medical and genetic factors. *eLife*, 8:e46179.

Tan TW, Shih CD, Concha-Moore KC, Diri MM, Hu B, Marrero D, Zhou W, and Armstrong DG (2019) Disparities in outcomes of patients admitted with diabetic foot infections. *PLoS One*, 14:e0211481. doi: 10.1371/journal.pone.0211481. eCollection .

Thal DR, Ronisz A, Tousseyn T, Rijal Upadhaya A, Balakrishnan K, Vandenberghe R, Vandebulcke M, von Arnim CAF, Otto M, Beach TG, Lilja J, Heurling K, Chakrabarty A, Ismail A, Buckley C, Smith APL, Kumar S, Farrar G, Walter J. Different aspects of Alzheimer's disease-related amyloid  $\beta$ -peptide pathology and their relationship to amyloid positron emission tomography imaging and dementia. *Acta Neuropathol Commun*. 2019 Nov 14;7(1):178. doi: 10.1186/s40478-019-0837-9. PubMed PMID: 31727169.

Thirunavu V, McCullough A, Su Y, Flores S, Dincer A, Morris JC, Cruchaga C, Benzinger TLS, Gordon BA. Higher Body Mass Index Is Associated with Lower Cortical Amyloid- $\beta$  Burden in Cognitively Normal Individuals in Late-Life. *J Alzheimers Dis*. 2019;69(3):817-827. doi: 10.3233/JAD-190154. PMID: 31156169

Toosizadeh N, Ehsani H, Wendel C, Zamrini E, Connor KO, Mohler J. Screening older adults for amnesic mild cognitive impairment and early-stage Alzheimer's disease using upper-extremity dual-tasking. *Sci Rep*. 2019 Jul 29;9(1):10911. doi:10.1038/s41598-019-46925-y. PubMed PMID: 31358792; PubMed Central PMCID: PMC6662814.

Torres VL, Vila-Castelar C, Bocanegra Y, Baena A, Guzmán-Vélez E, Aguirre-Acevedo DC, Tirado V, Munoz C, Henao E, Moreno S, Giraldo M, Acosta N, Rios Romenets S, Langbaum JB, Cho W, Reiman EM, Tariot PN, Rosselli M, Quiroz YT, Lopera F. Normative data stratified by age and education for a Spanish neuropsychological test battery: Results from the Colombian Alzheimer's prevention initiative registry. *Appl Neuropsychol Adult*. 2019 Jun 23:1-15. doi: 10.1080/23279095.2019.1627357. [Epub ahead of print] PMID: 31230490

Tsai PH, Kaur G, Gopalakrishna G. No MMSE for You! A Case of an "Uncooperative" Patient With Early-Onset Dementia. *Prim Care Companion CNS Disord*. 2019 Oct 24;21(5). pii: 19alz02544. doi: 10.4088/PCC.19alz02544. PMID: 31721488

van Dyck CH, Nygaard HB, Chen K, Donohue MC, Raman R, Rissman RA, Brewer JB, Koeppe RA, Chow TW, Rafii MS, Gessert D, Choi J, Turner RS, Kaye JA, Gale SA, Reiman EM, Aisen PS, Strittmatter SM. Effect of AZD0530 on Cerebral Metabolic Decline in Alzheimer Disease: A Randomized Clinical Trial. *JAMA Neurol*. 2019 Jul 22. doi: 10.1001/jamaneurol.2019.2050. [Epub ahead of print] PMID: 31329216

Varatharajah Y, Ramanan VK, Iyer R, Vemuri P; Alzheimer's Disease Neuroimaging Initiative. Predicting Short-term MCI-to-AD Progression Using Imaging, CSF, Genetic Factors, Cognitive Resilience, and Demographics. *Sci Rep*. 2019 Feb 19;9(1):2235. doi: 10.1038/s41598-019-38793-3. PMID: 30783207

Vaske OM, Bjork I, Salama SR, Beale H, Shah AT, Sanders L, Pfeil J, Lam DL, Learned K, Durbin A, Kephart ET, Currie R, Newton Y, Swatloski T, McColl D, Vivian J, Zhu J, Lee AG, Leung S, Spillinger A, Liu H, Liang WS, Byron SA, Berens ME, Resnick A, Lacayo N, Spunt S, Rangaswami A, Huynh V, Torno L, Plant A, Kirov I, Zabokrtsky K, Rassekh SR, Deyell R, Laskin J, Marra MA, Sender LS\*, Mueller S\*, Sweet-Cordero EA\*, Goldstein TC\*, Haussler D (2019) Comparative

tumor RNA sequencing analysis for difficult-to-treat pediatric and young adult patients with cancer. *JAMA Network Open* 2(10): e1913968; PMID: 31651965.

Walker DG, Lue LF, Beach TG, Tooyama I. Microglial Phenotyping in Neurodegenerative Disease Brains: Identification of Reactive Microglia with an Antibody to Variant of CD105/Endoglin. *Cells*. 2019 Jul 23;8(7). pii: E766. doi:10.3390/cells8070766. PubMed PMID: 31340569.

Wang Q, Wang Y, Liu J, Sutphen CL, Cruchaga C, Blazey T, Gordon BA, Su Y, Chen C, Shimony JS, Ances BM, Cairns NJ, Fagan AM, Morris JC, Benzinger TLS. Quantification of white matter cellularity and damage in preclinical and early symptomatic Alzheimer's disease. *Neuroimage Clin*. 2019;22:101767. doi: 10.1016/j.nicl.2019.101767. Epub 2019 Mar 13. PMID: 30901713

Wang Y, Hernandez G, Mack WJ, Schneider LS, Yin F, Brinton RD (2019) Retrospective analysis of phytoSERM for management of menopause--associated vasomotor symptoms and cognitive decline: a pilot study on pharmacogenomic effects of mitochondrial haplogroup and APOE genotype on therapeutic efficacy. *Menopause*, 27:57-65.

Wang, T, Yao J, Chen S, Mao J, and Brinton, RD (2019) Allopregnanolone Reverses Bioenergetic Deficits in Female Triple Transgenic Alzheimer's Mouse Model. *Neurotherapeutics*, doi: 10.1007/s13311-019-00793-6.

Webster THp, Phung T, Grande B, Karlins E, Richmond P, Couse M, Whitford W, and Wilson MA. 2019. Identifying, understanding, and correcting technical artifacts on the sex chromosomes in next-generation sequencing data. *GigaScience* 8(7):giz074.

Weinkauf C, Mazhar A, Vaishnav K., Hamadani AA, Cuccia DJ, and Armstrong, DG (2019) Near-instant noninvasive optical imaging of tissue perfusion for vascular assessment. *Journal of Vascular Surgery*, 69:555-562.

Wenjun Deng, C. X., Rob David, Diego Mastroeni, MingMing Ning, Eng H Lo, Paul D Coleman. *AmpliSeq Transcriptome of Laser Captured Neurons from Alzheimer Brain: Comparison of Single Cell Versus Neuron Pools*. *Aging and disease* (2019).

Wessels AM, Tariot PN, Zimmer JA, Selzler KJ, Bragg SM, Andersen SW, Landry J, Krull JH, Downing AM, Willis BA, Shcherbinin S, Mullen J, Barker P, Schumi J, Shering C, Matthews BR, Stern RA, Vellas B, Cohen S, MacSweeney E, Boada M, Sims JR. Efficacy and Safety of Lanabecestat for Treatment of Early and Mild Alzheimer Disease: The AMARANTH and DAYBREAK-ALZ Randomized Clinical Trials. *JAMA Neurol*. 2019 Nov 25. doi: 10.1001/jamaneurol.2019.3988. [Epub ahead of print] PMID: 31764959

White Z, Milad Nadia, Tehrani AY, Lamothe J, Hogg JC, Esfandiarei M, Seidman M, Booth S, Hackett TL, Morissette MC, Bernatchez P. Sildenafil prevents Marfan-associated emphysema and early pulmonary artery dilation in mice. *Am J Pathol*. 2019 Aug; 189(8):1536-1546.

Willeman, M.N., Chawla, M.K., Zempare, M.A., Biwer, L.A., Hoang, L.T., Uprety, A.R., Fitzhugh, M.C., De Both, M., Coleman, P.D., Trouard, T.P., Alexander, G.E., Mitchell, K.D., Barnes, C.A., Hale, T.M. & Huentelman, M. Gradual hypertension induction in middle-aged Cyp1a1-Ren2 transgenic rats produces significant impairments in spatial learning. *Physiol Rep* 7, e14010 (2019).

Wilson MA and Buetow KH. 2019. Novel mechanisms of cancer emerge when accounting for sex as a biological variable (accepted) *Cancer Research*. doi: 10.1158/0008-5472.CAN-19-2634

Wilson RC and Collins AGE (2019) Ten simple rules for the computational modeling of behavioral data. *eLife*, 8:e49547.

Wilson RC, Shenhav A, Straccia, M, and Cohen, JD (2019) The Eighty Five Percent Rule for Optimal Learning. *Nature Communications*, 10:4646. doi:10.1038/s41467-019-12552-4.

Wingo AP, Dammer EB, Breen MS, Logsdon BA, Duong DM, Troncosco JC, Thambisetty M, Beach TG, Serrano GE, Reiman EM, Caselli RJ, Lah JJ, Seyfried NT, Levey AI, Wingo TS. Large-scale proteomic analysis of human brain identifies proteins associated with cognitive trajectory in advanced age. *Nat Commun*. 2019 Apr 8;10(1):1619. doi: 10.1038/s41467-019-09613-z. PubMed PMID: 30962425.

Xu W, Bao P, Wang H, Wang T, Jiang X, Yang Y, Qin M, Wang R, Lorenzini I, Liao L, Sattler R, and Xu J. Reactivation of nonsense-mediated mRNA decay protects against C9orf72 dipeptide-repeat neurotoxicity. *Brain*, 2019 May 1;142(5):1349-1364. doi: 10.1093/brain/awz070

Xu, F., Ma, J., Sun, F., Lee, J., Coon, D., Xiao, Q., & Liang, Z. (2019). The sensitivity and specificity of The General Practitioner Assessment of Cognition in Chinese elders aged 80 and over. *American Journal of Alzheimer's Disease & Other Dementias*, 34, 523-529.

Yang Y, Chen Y, Chen K, Wei D, Li P, Zeng W, Pei J, Mao H, Jia J, Zhang Z. Increased intrinsic connectivity for structural atrophy and functional maintenance after acute ischaemic stroke. *Eur J Neurol*. 2019 Jun;26(6):935-942. doi: 10.1111/ene.13913. Epub 2019 Mar 12. PMID: 30681762

Yang Z, Zhuang X, Bird C, Sreenivasan K, Mishra V, Banks S, Cordes D; Alzheimer's Disease Neuroimaging Initiative. Performing Sparse Regularization and Dimension Reduction Simultaneously in Multimodal Data Fusion. *Front Neurosci*. 2019 Jul 3;13:642. doi: 10.3389/fnins.2019.00642. eCollection 2019. PMID: 31333396

Yanquez FJ, Peterson A, Weinkauff C, Goshima KR, Zhou W, Mohler J, Ehsani H, and Toosizadeh N (2019) Sensor-based upper-extremity frailty assessment for the vascular surgery risk stratification. *The Journal of Surgical Research*, 246:403-410.

Yonelinas AP, Rangnath CR, Ekstrom AD, and Wiltgen BL (2019) A Contextual Binding Theory of Episodic Memory: Systems Consolidation Reconsidered. *Nature Reviews Neuroscience*, 20: 364-375.

Yonelinas AP, Rangnath CR, Ekstrom AD, and Wiltgen BL (2019) Reply to 'Active and effective replay: Systems consolidation reconsidered again.' *Nature Reviews Neuroscience*, 20: 507-508.

Zabelina D, Friedman NP, and Andrews-Hanna JR (2019) Unity and diversity of executive functions in creativity. *Consciousness and Cognition*, 68: 47-56.

Zahr NM, Pohl KM, Saranathan M, Sullivan EV, and Pfefferbaum A. (2019) Hippocampal subfield CA2+3 exhibits accelerated aging in alcohol use disorder: A preliminary study. *Neuroimage Clinical* 22:101764.

Zahr NM, Sullivan EV, Pohl KM, Pfefferbaum A, and Saranathan M. (2019) Sensitivity of ventrolateral posterior thalamic nucleus to back pain in alcoholism and CDC nadir in HIV. *Human Brain Mapping*, <https://doi.org/10.1002/hbm.24880>.

Zhang YJ, Guo L, Gonzales PK, Gendron TF, Wu Y, Jansen-West K, O'Raw AD, Pickles SR, Prudencio M, Carlomagno Y, Gachechiladze MA, Ludwig C, Tian R, Chew J, DeTure M, Lin WL, Tong J, Daugherty LM, Yue M, Song Y, Andersen JW, Castanedes-Casey M, Kurti A, Datta A, Antognetti G, McCampbell A, Rademakers R, Oskarsson B, Dickson DW, Kampmann M, Ward ME, Fryer JD, Link CD, Shorter J, Petrucelli L. Heterochromatin anomalies and double-stranded RNA accumulation underlie C9orf72 poly(PR) toxicity. *Science*. 2019 Feb 15;363(6428). pii: eaav2606. doi: 10.1126/science.aav2606. PMID: 30765536 Free PMC Article

Zhao Q, Liu M, Ha L, Zhou Y; Alzheimer's Disease Neuroimaging Initiative. Quantitative <sup>18</sup>F-AV1451 Brain Tau PET Imaging in Cognitively Normal Older Adults, Mild Cognitive Impairment, and Alzheimer's Disease Patients. *Front Neurol*. 2019 May 15;10:486. doi: 10.3389/fneur.2019.00486. eCollection 2019. PMID: 31156534

Zhou X, Chen Y, Mok KY, Kwok TCY, Mok VCT, Guo Q, Ip FC, Chen Y, Mullanpudi N; Alzheimer's Disease Neuroimaging Initiative, Giusti-Rodríguez P, Sullivan PF, Hardy J, Fu AKY, Li Y, Ip NY. Non-coding variability at the APOE locus contributes to the Alzheimer's risk. *Nat Commun*. 2019 Jul 25;10(1):3310. doi: 10.1038/s41467-019-10945-z. PMID: 31346172

Zhu H, Wang W, Li H, Chen K, Li P, Li X, Zhang J, Wei D, Chen Y. Basal Ganglia-Cortical Circuit Disruption in Subcortical Silent Lacunar Infarcts. *Front Neurol*. 2019 Jun 25;10:660. doi: 10.3389/fneur.2019.00660. eCollection 2019. PMID: 31293502

## 2020 Publications and Manuscripts

Acevedo-Molina MC, Matijevic S, Grilli MD. Beyond episodic remembering: elaborative retrieval of lifetime periods in young and older adults. *Memory*. 2020 Jan;28(1):83-93. doi: 10.1080/09658211.2019.1686152. Epub 2019 Oct 31. PubMed PMID: 31665972; PubMed Central PMCID: PMC6934901.

Al Rubaye, H, Adamson, CC, Jadavji, NM. The role of maternal diet on offspring gut microbiota development: A review. *J Neuro Res*. 2020; 00: 1– 10.

Alber J, Goldfarb D, Thompson LI, Arthur E, Hernandez K, Cheng D, DeBuc DC, Cordeiro F, Provetti-Cunha L, den Haan J, Van Stavern GP, Salloway SP, Sinoff S, Snyder PJ. Developing retinal biomarkers for the earliest stages of Alzheimer's disease: What we know, what we don't, and how to move forward. *Alzheimers Dement*. 2020 Jan;16(1):229-243. doi: 10.1002/alz.12006. Review. PMID: 31914225

Alber J, Maruff P, Santos CY, Ott BR, Salloway SP, Yoo DC, Noto RB, Thompson LI, Goldfarb D, Arthur E, Song A, Snyder PJ. Disruption of cholinergic neurotransmission, within a cognitive challenge paradigm, is indicative of A $\beta$ -related cognitive impairment in preclinical Alzheimer's disease after a 27-month delay interval. *Alzheimers Res Ther*. 2020 Mar 24;12(1):31. doi: 10.1186/s13195-020-00599-1. PMID: 32209123

Andrews-Hanna J, Christoff K, and O'Connor MF. (in press) Dynamic regulation of internal experience. In: R Lane, L Ryan, and L Nadel (Eds.). *The Neuroscience of Enduring Change: The Neural Basis of Talk Therapies*. New York, NY: Oxford University Press U.S.A.

Andrews-Hanna JR, Fox KC, Irving Z, Spreng RN, and Christoff K. (in press) The neuroscience of spontaneous thought: An evolving, interdisciplinary field. In: *The Oxford Handbook of Spontaneous Thought: Mind-wandering, Creativity, & Dreaming*. New York City: Oxford University Press.

Bai B, Wang X, Li Y, Chen PC, Yu K, Dey KK, Yarbro JM, Han X, Lutz BM, Rao S, Jiao Y, Sifford JM, Han J, Wang M, Tan H, Shaw TI, Cho JH, Zhou S, Wang H, Niu M, Mancieri A, Messler KA, Sun X, Wu Z, Pagala V, High AA, Bi W, Zhang H, Chi H, Haroutunian V, Zhang B, Beach TG, Yu G, Peng J. Deep Multilayer Brain Proteomics Identifies Molecular Networks in Alzheimer's Disease Progression. *Neuron*. 2020 Mar 18;105(6):975-991.e7. doi: 10.1016/j.neuron.2019.12.015. Epub 2020 Jan 8. PubMed PMID: 31926610.

Barnes, C.A., Ryan, L. and Peterson, M.A. (2020), Nadel special issue introduction. *Hippocampus*. doi:10.1002/hipo.23176.

Bartoň M, Fňášková M, Rektorová I, Mikl M, Mareček R, Rapcsak SZ, Rektor I. The role of the striatum in visuomotor integration during handwriting: an fMRI study. *J Neural Transm (Vienna)*. 2020 Mar;127(3):331-337. doi: 10.1007/s00702-019-02131-8. Epub 2020 Jan 4. PubMed PMID: 31901984.

Bassiouni A, Paramasivan S, Shiffer A, Dillon MR, Cope EK, Cooksley C, Ramezanpour M, Moraitis S, Ali MJ, Bleier BS, Callejas C, Cornet ME, Douglas RG, Dutra D, Georgalas C, Harvey RJ, Hwang PH, Luong AU, Schlosser RJ, Tantilipikorn P, Tewfik MA, Vreugde S, Wormald PJ, Caporaso JG, Psaltis AJ. Microbiotyping the Sinonasal Microbiome. *Front Cell Infect Microbiol*.

2020 Apr 8;10:137. doi: 10.3389/fcimb.2020.00137. eCollection 2020. PubMed PMID: 32322561; PubMed CentralPMCID: PMC7156599.

Beach TG, Adler CH, Zhang N, Serrano GE, Sue LI, Driver-Dunckley E, Mehta SH, Zamrini EE, Sabbagh MN, Shill HA, Belden CM, Shprecher DR, Caselli RJ, Reiman EM, Davis KJ, Long KE, Nicholson LR, Intorcchia AJ, Glass MJ, Walker JE, Callan MM, Oliver JC, Arce R, Gerkin RC. Severe hyposmia distinguishes neuropathologically confirmed dementia with Lewy bodies from Alzheimer's disease dementia. *PLoS One*. 2020 Apr 22;15(4):e0231720. doi: 10.1371/journal.pone.0231720. eCollection 2020. PubMed PMID: 32320406; PubMed Central PMCID: PMC7176090.

Beach, TG., Malek-Ahmadi, M. "Alzheimer's Disease Neuropathological Comorbidities Are Common in the Younger-Old." *bioRxiv* (2020): 2020.01.03.894451. Web. 06 April. 2020 (submitted)

Beitchman JA, Griffiths DR, Hur Y, Ogle SB, Bromberg CE, Morrison HW, Lifshitz J, Adelson PD, Currier Thomas T. Corrigendum: Experimental Traumatic Brain Injury Induces Chronic Glutamatergic Dysfunction in Amygdala Circuitry Known to Regulate Anxiety-Like Behavior. *Front Neurosci*. 2020 Mar 13;14:212. doi: 10.3389/fnins.2020.00212. eCollection 2020. PubMed PMID: 32231519; PubMed Central PMCID: PMC7083151.

Bell LC, Stokes AM, Quarles CC. Analysis of postprocessing steps for residue function dependent dynamic susceptibility contrast (DSC)-MRI biomarkers and their clinical impact on glioma grading for both 1.5 and 3T. *J Magn Reson Imaging*. 2020 Feb;51(2):547-553. doi: 10.1002/jmri.26837. Epub 2019 Jun 17. PubMed PMID: 31206948.

Belser-Ehrlich J, Adrian Lafo J, Mangal P, Bradley M, Wicklund M, Bowers D. Neurocognitive profile of a man with Dandy-Walker malformation: Evidence of subtle cerebellar cognitive affective syndrome. *Clin Neuropsychol*. 2020 Apr;34(3):591-610. Epub 2019 Mar 1. PubMed PMID: 30821610.

Bharadwaj, VN, Copeland, C, Mathew, E, Newbern, J, Anderson, TR, Lifshitz, J, Kodibagkar, VD, Stabenfeldt, SE. Sex-dependent macromolecule and nanoparticle delivery in experimental brain injury. In Review. (posted in *bioRxiv*. 2019; 817296; DOI: 10.1101/817296)

Borazanci E, Korn R, Liang WS, Guarnieri C, Haag S, Snyder C, Hendrickson K, Caldwell L, Von Hoff D, Jameson G. An Analysis of Patients with DNA Repair Pathway Mutations Treated with a PARP Inhibitor. *Oncologist*. 2020 Jan;25(1):e60-e67. doi: 10.1634/theoncologist.2018-0905. Epub 2019 Aug 7. PubMed PMID: 31391296; PubMed Central PMCID: PMC6964119

Borden ES, Kang P, Natri HM, Phung TN, Wilson MA, Buetow KH, Hastings KT. Neoantigen Fitness Model Predicts Lower Immune Recognition of Cutaneous Squamous Cell Carcinomas Than Actinic Keratoses. *Front Immunol*. 2019 Nov 29;10:2799. doi: 10.3389/fimmu.2019.02799. eCollection 2019. PubMed PMID: 31849976; PubMed Central PMCID: PMC6896054.

Branigan GL, Soto M, Neumayer L, Rodgers K, Brinton RD. Association Between Hormone-Modulating Breast Cancer Therapies and Incidence of Neurodegenerative Outcomes for Women With Breast Cancer. *JAMA Netw Open*. 2020 Mar 2;3(3):e201541. doi: 10.1001/jamanetworkopen.2020.1541. PubMed PMID: 32207833; PubMed Central PMCID: PMC7093781.

Brookhouser N, Tekel SJ, Standage-Beier K, Schwarz G, Nguyen T, Wang X, Brafman DA. BIG-TREE: Base-edited isogenic hiPSC line generation using a transient reporter for editing enrichment. *Stem Cell Reports*. 2020 Feb 11;14(2):1-8.

Caselli RJ, Langlais BT, Dueck AC, Chen Y, Su Y, Locke DEC, Woodruff BK, Reiman EM. Neuropsychological decline up to 20 years before incident mild cognitive impairment. *Alzheimers Dement*. 2020 Mar;16(3):512-523. doi: 10.1016/j.jalz.2019.09.085. Epub 2020 Jan 6. PMID: 31787561

Chou YH, Ton That V, Chen AY, Sundman M, Huang YZ. TMS-induced seizure cases stratified by population, stimulation protocol, and stimulation site: A systematic literature search. *Clin Neurophysiol*. 2020 May;131(5):1019-1020. doi: 10.1016/j.clinph.2020.02.008. Epub 2020 Mar 3. PubMed PMID: 32193163.

Conrado DJ, Duvvuri S, Geerts H, Burton J, Biesdorf C, Ahamadi M, Macha S, Hather G, Morales JF, Podichetty J, Nicholas T, Stephenson D, Trame M, Romero K, Corrigan B; Drug Development Tools in the Alzheimer Disease Continuum (DDT-AD) Working Group. "Challenges in Alzheimer Disease Drug Discovery and Development: The Role of Modeling, Simulation and Open Data." *Clin Pharmacol Ther*, January 18, 2020.

Conrado, DJ., Burton J, Hill D, Willis B, Sinha V, Stone J, Coello N, Wang W, Chen D, Nicholas T, Gold M, Hartley E, Kern VD, Romero K for the Alzheimer's Disease Neuroimaging Initiative for the Critical Path for Alzheimer's Disease (CPAD) "Hippocampal Neuroimaging-Informed Clinical Trial Enrichment Tool for Amnesic Mild Cognitive Impairment Using Open Data." *Clinical Pharmacology and Therapeutics*, January 3, 2020.

Deepti Lall, Ileana Lorenzini, Thomas Mahan, Jason D. Ulrich, Hayk Davytan, Jessica Rexach, A.K.M Ghulam Mohammad, Thomas A. Mota, Jesse Landeros, Junwon Kim, Layla Ghaffari, Shaughn Bell, Daniel H. Geshwind, Mathew Blurton-Jones, David M. Holtzman, Rita Sattler, Robert H. Baloh. C9orf72 deficiency promotes microglial mediated synaptic loss in aging and amyloid accumulation. *Nature Neuroscience*, under review.

Ehsani H, Parvaneh S, Mohler J, Wendel C, Zamrini E, O'Connor K, Toosizadeh N. Can motor function uncertainty and local instability within upper-extremity dual-tasking predict amnesic mild cognitive impairment and early-stage Alzheimer's disease? *Comput Biol Med*. 2020 Mar 19;120:103705. doi:10.1016/j.compbio.2020.103705. [Epub ahead of print] PubMed PMID: 32217286

Ekstrom AD, Harootyan SK, Huffman DJ. Grid coding, spatial representation, and navigation: Should we assume an isomorphism? *Hippocampus*. 2020 Apr;30(4):422-432. doi: 10.1002/hipo.23175. Epub 2019 Nov 18. PubMed PMID: 31742364.

Fleisher AS, Pontecorvo MJ, Devous MD Sr, Lu M, Arora AK, Trucchio SP, Aldea P, Flitter M, Locascio T, Devine M, Siderowf A, Beach TG, Montine TJ, Serrano GE, Curtis C, Perrin A, Salloway S, Daniel M, Wellman C, Joshi AD, Irwin DJ, Lowe VJ, Seeley WW, Ikonomic MD, Masdeu JC, Kennedy I, Harris T, Navitsky M, Southekal S, Mintun MA; A16 Study Investigators. Positron Emission Tomography Imaging With [18F]flortaucipir and Postmortem Assessment of Alzheimer Disease Neuropathologic Changes. *JAMA Neurol*. 2020 Apr 27. doi: 10.1001/jamaneurol.2020.0528. [Epub ahead of print] PubMed PMID: 32338734; PubMed Central PMCID: PMC7186920.

Franzmeier N, Neitzel J, Rubinski A, Smith R, Strandberg O, Ossenkoppele R, Hansson O, Ewers M; Alzheimer's Disease Neuroimaging Initiative (ADNI). Functional brain architecture is associated with the rate of tau accumulation in Alzheimer's disease. *Nat Commun.* 2020 Jan 17;11(1):347. doi: 10.1038/s41467-019-14159-1. PMID: 31953405

Gatchel JR, Lopera F, Norton DJ, Baena A, Guzman-Velez E, Sanchez JS, d'Oleire Uquillas F, Schultz A, Vannini P, Artola A, Amariglio RE, Rentz DM, Tariot PN, Reiman EM, Johnson KA, Sperling RA, Marshall GA, Quiroz YT. Association of subjective cognitive decline with markers of brain pathology in preclinical autosomal dominant Alzheimer's disease. *J Neurol Neurosurg Psychiatry.* 2020 Mar;91(3):330-332. doi: 10.1136/jnnp-2019-321205. Epub 2019 Dec 24. PMID: 31874859

Ghisays V, Goradia DD, Protas H, Bauer RJ 3rd, Devadas V, Tariot PN, Lowe VJ, Knopman DS, Petersen RC, Jack CR Jr, Caselli RJ, Su Y, Chen K, Reiman EM. Brain imaging measurements of fibrillar amyloid- $\beta$  burden, paired helical filament tau burden, and atrophy in cognitively unimpaired persons with two, one, and no copies of the APOE  $\epsilon$ 4 allele. *Alzheimers Dement.* 2020 Jan 16. doi: 10.1016/j.jalz.2019.08.195. [Epub ahead of print] PMID: 31831374

Giordano KR, Denman CR, Dollish HK, Fernandez F, Lifshitz J, Akhter M, Rowe RK. Intracerebral hemorrhage in the mouse altered sleep-wake patterns and activated microglia. *Exp Neurol.* 2020 May;327:113242. doi: 10.1016/j.expneurol.2020.113242. Epub 2020 Feb 11. PubMed PMID: 32057796.

Grasby KL, Jahanshad N, Painter JN, Colodro-Conde L, Bralten J, Hibar DP, Lind PA, Pizzagalli F, Ching CRK, McMahon MAB, Shatokhina N, Zsembik LCP, Thomopoulos SI, Zhu AH, Strike LT, Agartz I, Alhusaini S, Almeida MAA, Alnæs D, Amlien IK, Andersson M, Ard T, Armstrong NJ, Ashley-Koch A, Atkins JR, Bernard M, Brouwer RM, Buimer EEL, Bülow R, Bürger C, Cannon DM, Chakravarty M, Chen Q, Cheung JW, Couvy-Duchesne B, Dale AM, Dalvie S, de Araujo TK, de Zubicaray GI, de Zwarte SMC, den Braber A, Doan NT, Dohm K, Ehrlich S, Engelbrecht HR, Erk S, Fan CC, Fedko IO, Foley SF, Ford JM, Fukunaga M, Garrett ME, Ge T, Giddaluru S, Goldman AL, Green MJ, Groenewold NA, Grotegerd D, Gurholt TP, Gutman BA, Hansell NK, Harris MA, Harrison MB, Haswell CC, Hauser M, Herms S, Heslenfeld DJ, Ho NF, Hoehn D, Hoffmann P, Holleran L, Hoogman M, Hottenga JJ, Ikeda M, Janowitz D, Jansen IE, Jia T, Jockwitz C, Kanai R, Karama S, Kasperaviciute D, Kaufmann T, Kelly S, Kikuchi M, Klein M, Knapp M, Knodt AR, Krämer B, Lam M, Lancaster TM, Lee PH, Lett TA, Lewis LB, Lopes-Cendes I, Luciano M, Macciardi F, Marquand AF, Mathias SR, Melzer TR, Milaneschi Y, Mirza-Schreiber N, Moreira JCV, Mühleisen TW, Müller-Myhsok B, Najt P, Nakahara S, Nho K, Olde Loohuis LM, Orfanos DP, Pearson JF, Pitcher TL, Pütz B, Quidé Y, Ragothaman A, Rashid FM, Reay WR, Redlich R, Reinbold CS, Repple J, Richard G, Riedel BC, Risacher SL, Rocha CS, Mota NR, Salminen L, Saremi A, Saykin AJ, Schlag F, Schmaal L, Schofield PR, Secolin R, Shapland CY, Shen L, Shin J, Shumskaya E, Sønderby IE, Sprooten E, Tansey KE, Teumer A, Thalamuthu A, Tordesillas-Gutiérrez D, Turner JA, Uhlmann A, Vallerga CL, van der Meer D, van Donkelaar MMJ, van Eijk L, van Erp TGM, van Haren NEM, van Rooij D, van Tol MJ, Veldink JH, Verhoef E, Walton E, Wang M, Wang Y, Wardlaw JM, Wen W, Westlye LT, Whelan CD, Witt SH, Wittfeld K, Wolf C, Wolfers T, Wu JQ, Yasuda CL, Zaremba D, Zhang Z, Zwiers MP, Artiges E, Assareh AA, Ayesa-Arriola R, Belger A, Brandt CL, Brown GG, Cichon S, Curran JE, Davies GE, Degenhardt F, Dennis MF, Dietsche B, Djurovic S, Doherty CP, Espiritu R, Garijo D, Gil Y, Gowland PA, Green RC, Häusler AN, Heindel W, Ho BC, Hoffmann WU, Holsboer F, Homuth G, Hosten N, Jack CR Jr, Jang M, Jansen A, Kimbrel NA, Kolskár K, Koops S, Krug A, Lim KO, Luyck JJ, Mathalon DH, Mather KA, Mattay VS, Matthews S, Mayoral Van Son J, McEwen SC, Melle I, Morris DW, Mueller BA, Nauck M, Nordvik JE, Nöthen MM, O'Leary DS, Opel N, Martinot

MP, Pike GB, Preda A, Quinlan EB, Rasser PE, Ratnakar V, Reppermund S, Steen VM, Tooney PA, Torres FR, Veltman DJ, Voyvodic JT, Whelan R, White T, Yamamori H, Adams HHH, Bis JC, Debette S, Decarli C, Fornage M, Gudnason V, Hofer E, Ikram MA, Launer L, Longstreth WT, Lopez OL, Mazoyer B, Mosley TH, Roshchupkin GV, Satizabal CL, Schmidt R, Seshadri S, Yang Q; Alzheimer's Disease Neuroimaging Initiative; CHARGE Consortium; EPIGEN Consortium; IMAGEN Consortium; SYS Consortium; Parkinson's Progression Markers Initiative, Alvim MKM, Ames D, Anderson TJ, Andreassen OA, Arias-Vasquez A, Bastin ME, Baune BT, Beckham JC, Blangero J, Boomsma DI, Brodaty H, Brunner HG, Buckner RL, Buitelaar JK, Bustillo JR, Cahn W, Cairns MJ, Calhoun V, Carr VJ, Caseras X, Caspers S, Cavalleri GL, Cendes F, Corvin A, Crespo-Facorro B, Dalrymple-Alford JC, Dannlowski U, de Geus EJC, Deary IJ, Delanty N, Depondt C, Desrivieres S, Donohoe G, Espeseth T, Fernández G, Fisher SE, Flor H, Forstner AJ, Francks C, Franke B, Glahn DC, Gollub RL, Grabe HJ, Gruber O, Håberg AK, Hariri AR, Hartman CA, Hashimoto R, Heinz A, Henskens FA, Hillegers MHJ, Hoekstra PJ, Holmes AJ, Hong LE, Hopkins WD, Hulshoff Pol HE, Jernigan TL, Jönsson EG, Kahn RS, Kennedy MA, Kircher TTJ, Kochunov P, Kwok JBJ, Le Hellard S, Loughland CM, Martin NG, Martinot JL, McDonald C, McMahon KL, Meyer-Lindenberg A, Michie PT, Morey RA, Mowry B, Nyberg L, Oosterlaan J, Ophoff RA, Pantelis C, Paus T, Pausova Z, Penninx BWJH, Polderman TJC, Posthuma D, Rietschel M, Roffman JL, Rowland LM, Sachdev PS, Sämann PG, Schall U, Schumann G, Scott RJ, Sim K, Sisodiya SM, Smoller JW, Sommer IE, St Pourcain B, Stein DJ, Toga AW, Trollor JN, Van der Wee NJA, van 't Ent D, Völzke H, Walter H, Weber B, Weinberger DR, Wright MJ, Zhou J, Stein JL, Thompson PM, Medland SE; Enhancing Neuroimaging Genetics through Meta-Analysis Consortium (ENIGMA)—Genetics working group. The genetic architecture of the human cerebral cortex. *Science*. 2020 Mar 20;367(6484). pii: eaay6690. doi: 10.1126/science.aay6690. PMID: 32193296

Gray DT, Umapathy L, De La Peña NM, Burke SN, Engle JR, Trouard TP, and Barnes CA (2020) Auditory processing deficits are selectively associated with medial temporal lobe mnemonic function and white matter integrity in aging macaques. *Cerebral Cortex*, doi: 10.1093/cercor/bhz275. [Epub ahead of print]

Grilli MD and Ryan L. (in press) Autobiographical memory and the self-concept. In: R Lane, L Ryan, and L Nadel (Eds.). *The Neuroscience of Enduring Change: The Neural Basis of Talk Therapies*. New York, NY: Oxford University Press U.S.A.

Hejtmanek L, Starrett M, Ferrer E, Ekstrom AD. How Much of What We Learn in Virtual Reality Transfers to Real-World Navigation? *Multisens Res*. 2020 Jan 21:1-25. doi: 10.1163/22134808-20201445. [Epub ahead of print] PubMed PMID: 31972540.

Huentelman, M.J., Talboom, J.S., Lewis, C.R., Chen, Z., Barnes, C.A. (2020) Reinventing neuroaging research in the digital age. *Trends in Neuroscience*, 43:17–23.

Ilori TO, Viera E, Wilson J, Moreno F, Menon U, Ehiri J, Peterson R, Vemulapalli T, StimsonRiahi SC, Rosales C, Calhoun E, Sokan A, Karnes JH, Reiman E, Ojo A, Theodorou A, Ojo T. Approach to High Volume Enrollment in Clinical Research: Experiences from an All of Us Research Program Site. *Clin Transl Sci*. 2020 Jan 31. doi: 10.1111/cts.12759. [Epub ahead of print] PMID: 32004412

Itzhaki RF, Golde TE, Heneka MT, Readhead B. Do infections have a role in the pathogenesis of Alzheimer disease? *Nat Rev Neurol*. 2020 Apr;16(4):193-197. doi: 10.1038/s41582-020-0323-9. Epub 2020 Mar 9. Review. PubMed PMID: 32152461

Janelidze S, Mattsson N, Palmqvist S, Smith R, Beach TG, Serrano GE, Chai X, Proctor NK, Eichenlaub U, Zetterberg H, Blennow K, Reiman EM, Stomrud E, Dage JL, Hansson O. Plasma P-tau181 in Alzheimer's disease: relationship to other biomarkers, differential diagnosis, neuropathology and longitudinal progression to Alzheimer's dementia. *Nat Med.* 2020 Mar;26(3):379-386. doi: 10.1038/s41591-020-0755-1. Epub 2020 Mar 2. PubMed PMID: 32123385.

Jiang H, Lu N, Chen K, Yao L, Li K, Zhang J, Guo X. Predicting Brain Age of Healthy Adults Based on Structural MRI Parcellation Using Convolutional Neural Networks. *Front Neurol.* 2020 Jan 8;10:1346. doi: 10.3389/fneur.2019.01346. eCollection 2019. PMID: 31969858

Johnson ECB, Dammer EB, Duong DM, Ping L, Zhou M, Yin L, Higginbotham LA, Guajardo A, White B, Troncoso JC, Thambisetty M, Montine TJ, Lee EB, Trojanowski JQ, Beach TG, Reiman EM, Haroutunian V, Wang M, Schadt E, Zhang B, Dickson DW, Ertekin-Taner N, Golde TE, Petyuk VA, De Jager PL, Bennett DA, Wingo TS, Rangaraju S, Hajjar I, Shulman JM, Lah JJ, Levey AI, Seyfried NT. Large-scale proteomic analysis of Alzheimer's disease brain and cerebrospinal fluid reveals early changes in energy metabolism associated with microglia and astrocyte activation. *Nat Med.* 2020 Apr 13. doi: 10.1038/s41591-020-0815-6. [Epub ahead of print] PubMed PMID: 32284590.

Kalani MYS, Alsop E, Meechoovet B, Beecroft T, Agrawal K, Whitsett TG, Huentelman MJ, Spetzler RF, Nakaji P, Kim S, Van Keuren-Jensen K. Extracellular microRNAs in blood differentiate between ischaemic and haemorrhagic stroke subtypes. *J Extracell Vesicles.* 2020 Jan 24;9(1):1713540. doi: 10.1080/20013078.2020.1713540. eCollection 2020. PubMed PMID: 32128071; PubMed Central PMCID: PMC7034450.

Karamanova N, Truran S, Serrano GE, Beach TG, Madine J, Weissig V, Davies HA, Veldhuizen J, Nikkiah M, Hansen M, Zhang W, D'Souza K, Franco DA, Migrino RQ. Endothelial Immune Activation by Medin: Potential Role in Cerebrovascular Disease and Reversal by Monosialoganglioside-Containing Nanoliposomes. *J Am Heart Assoc.* 2020 Jan 21;9(2):e014810. doi: 10.1161/JAHA.119.014810. Epub 2020 Jan 13. PubMed PMID: 31928157.

Kari E, Llaci L, Go JL, Naymik M, Knowles JA, Leal SM, Rangasamy S, Huentelman MJ, Liang W, Friedman RA, Schrauwen I. Genes Implicated in Rare Congenital Inner Ear and Cochleovestibular Nerve Malformations. *Ear Hear.* 2020 Jan 24. doi: 10.1097/AUD.0000000000000819. [Epub ahead of print] PubMed PMID: 31985533.

Karim R, Dang HM, Hodis HN, Stanczyk FZ, Brinton RD, and Mack MJ (2020) Association of hot flushes with ghrelin and adipokines in early versus late postmenopausal women. *Menopause*, doi: 10.1097/GME.0000000000001508. [Epub ahead of print]

Kaufman JA, Castro MJ, Ruiz SA, Jentarra GM, Chavira B, Rodriguez-Sosa JR. (2020). Clearing, immunofluorescence, and confocal microscopy for the three-dimensional imaging of murine testes and study of testis biology. *Journal of Structural Biology*, March 1, 209(3):107449. doi: 10.1016/j.jsb.2020.

Keehoon Lee, Shari Kyman, Oliver Kask, Emily K Cope. Role of *Malassezia* in chronic rhinosinusitis. Submitted. 2020.

Knox MG, Adler CH, Shill HA, Driver-Dunckley E, Mehta SA, Belden C, Zamrini E, Serrano G, Sabbagh MN, Caviness JN, Sue LI, Davis KJ, Dugger BN, Beach TG. Neuropathological Findings

in Parkinson's Disease With Mild Cognitive Impairment. *Mov Disord.* 2020 Feb 8. doi: 10.1002/mds.27991. [Epub ahead of print] PubMed PMID: 32034933.

Kraemer C, Nisson P, Wheeler G, Guzmán Pérez-Carrillo GJ, Bernstein A, Hsu CH, Bock D, Trouard T, Zhou W. Patient risk factors associated with embolic stroke volumes following revascularization. *J Vasc Surg.* 2020 Apr 3. pii: S0741-5214(20)30467-5. doi: 10.1016/j.jvs.2020.02.040. [Epub ahead of print] PubMed PMID: 32251775.

Lawrence AV, Cardoza J, Ryan L. Medial temporal lobe regions mediate complex visual discriminations for both objects and scenes: A process-based view. *Hippocampus.* 2020 Mar 12. doi: 10.1002/hipo.23203. [Epub ahead of print] PubMed PMID: 32163223.

Lester AW, Kapellusch AJ, Barnes CA. A novel apparatus for assessing visual cue-based navigation in rodents. *J Neurosci Methods.* 2020 May 15;338:108667. doi: 10.1016/j.jneumeth.2020.108667. Epub 2020 Mar 10. PubMed PMID: 32169584.

Lewis CR, Taguinod F, Jepsen WM, Cohen J, Agrawal K, Huentelman MJ, Smith CJ, Ringenbach SDR, Braden BB. Telomere Length and Autism Spectrum Disorder Within the Family: Relationships With Cognition and Sensory Symptoms. *Autism Res.* 2020 Apr 23. doi: 10.1002/aur.2307. [Epub ahead of print] PubMed PMID: 32323911

Li X, Wang Y, Wang W, Huang W, Chen K, Xu K, Zhang J, Chen Y, Li H, Wei D, Shu N, Zhang Z. Age-Related Decline in the Topological Efficiency of the Brain Structural Connectome and Cognitive Aging. *Cereb Cortex.* 2020 Mar 26. pii: bhaa066. doi: 10.1093/cercor/bhaa066. [Epub ahead of print] PMID: 32219315

Li X, Xia J, Ma C, Chen K, Xu K, Zhang J, Chen Y, Li H, Wei D, Zhang Z. Accelerating Structural Degeneration in Temporal Regions and Their Effects on Cognition in Aging of MCI Patients. *Cereb Cortex.* 2020 Jan 10;30(1):326-338. doi: 10.1093/cercor/bhz090. PMID: 31169867

Liu Z, Dai X, Zhang H, Shi R, Hui Y, Jin X, Zhang W, Wang L, Wang Q, Wang D, Wang J, Tan X, Ren B, Liu X, Zhao T, Wang J, Pan J, Yuan T, Chu C, Lan L, Yin F, Cadenas E, Shi L, Zhao S, Liu X. (2020) Gut microbiota mediates intermittent-fasting alleviation of diabetes-induced cognitive impairment. *Nature Communications* 11:855. doi: 10.1038/s41467-020-14676-4.

Malek-Ahmadi M, Perez SE, Chen K, Mufson EJ. Braak Stage, Cerebral Amyloid Angiopathy, and Cognitive Decline in Early Alzheimer's Disease. *J Alzheimers Dis.* 2020;74(1):189-197. doi: 10.3233/JAD-191151. PubMed PMID: 31985469.

Matuskey D, Tinaz S, Wilcox KC, Naganawa M, Toyonaga T, Dias M, Henry S, Pittman B, Ropchan J, Nabulsi N, Suridjan I, Comley RA, Huang Y, Finnema SJ, Carson RE Synaptic changes in Parkinson's disease assessed with in-vivo imaging. (PMID 31953875) *Ann Neurol.* 2020 Mar;87(3):329-338. doi: 10.1002/ana.25682. Epub 2020 Feb 5.

Mi Y, Qi G, Brinton RD, Yin F. Mitochondria-Targeted Therapeutics for Alzheimer's Disease: The Good, the Bad, the Potential. *Antioxid Redox Signal.* 2020 Apr 21. doi: 10.1089/ars.2020.8070. [Epub ahead of print] PubMed PMID: 32143551.

Miotto EC, Balardin JB, Martin MDGM, Polanczyk GV, Savage CR, Miguel EC, Batistuzzo MC. Effects of semantic categorization strategy training on episodic memory in children and

adolescents. PLoS One. 2020 Feb 18;15(2):e0228866. doi: 10.1371/journal.pone.0228866. eCollection 2020. PMID: 32069310

Montagne A, Nation DA, Sagare AP, Barisano G, Sweeney MD, Chakhoyan A, Pachicano M, Joe E, Nelson AR, D'Orazio LM, Buennagel DP, Harrington MG, Benzinger TLS, Fagan AM, Ringman JM, Schneider LS, Morris JC, Reiman EM, Caselli RJ, Chui HC, Tcw J, Chen Y, Pa J, Conti PS, Law M, Toga AW, Zlokovic BV. APOE4 leads to blood-brain barrier dysfunction predicting cognitive decline. *Nature*. 2020 May;581(7806):71-76. doi: 10.1038/s41586-020-2247-3. Epub 2020 Apr 29. PubMed PMID: 32376954.

Moore S, Rabichow B, Sattler R. Hitchhiker's Guide to Nucleocytoplasmic Trafficking in Neurodegeneration. *Neurochemical Research*. 2020 Feb 21. doi: 10.1007/s11064-020-02989-1. [Epub ahead of print] PubMed PMID: 32086712.

Moreno-Rodriguez M, Perez SE, Nadeem M, Malek-Ahmadi M, Mufson EJ. Frontal cortex chitinase and pentraxin neuroinflammatory alterations during the progression of Alzheimer's disease. *J Neuroinflammation*. 2020 Feb 17;17(1):58. doi: 10.1186/s12974-020-1723-x. PubMed PMID: 32066474

Oliver Kask, Shari Kyman, Arron Shiffer, Jay Sutcliffe, Dierdra Byucara, J Gregory Caporaso, Emily K. Cope. Temporal Stability of the Athlete Nasal Microbiome Under Distinct Environmental Pressures. Submitted. 2020. Pre-print.

Orme T, Hernandez D, Ross OA, Kun-Rodrigues C, Darwent L, Shepherd CE, Parkkinen L, Ansoorge O, Clark L, Honig LS, Marder K, Lemstra A, Rogaeva E, St George-Hyslop P, Londos E, Zetterberg H, Morgan K, Troakes C, Al-Sarraj S, Lashley T, Holton J, Compta Y, Van Deerlin V, Trojanowski JQ, Serrano GE, Beach TG, Lesage S, Galasko D, Masliah E, Santana I, Pastor P, Tienari PJ, Myllykangas L, Oinas M, Revesz T, Lees A, Boeve BF, Petersen RC, Ferman TJ, Escott-Price V, Graff-Radford N, Cairns NJ, Morris JC, Pickering-Brown S, Mann D, Halliday G, Stone DJ, Dickson DW, Hardy J, Singleton A, Guerreiro R, Bras J. Analysis of neurodegenerative disease-causing genes in dementia with Lewy bodies. *Acta Neuropathol Commun*. 2020 Jan 29;8(1):5. PubMed PMID: 31996268.

Ornelas AS, Adler CH, Serrano GE, Curry JR, Shill HA, Kopyov O, Beach TG. Co-Existence of tau and  $\alpha$ -synuclein pathology in fetal graft tissue at autopsy: A case report. *Parkinsonism Relat Disord*. 2020 Feb;71:36-39. doi: 10.1016/j.parkreldis.2019.12.013. Epub 2020 Jan 11. PubMed PMID: 32004818; PubMed Central PMCID: PMC7085430.

Ortiz JB, Sukhina A, Balkan B, Harootunian G, Adelson PD, Lewis KS, Oatman O, Subbian V, Rowe RK, Lifshitz J. Epidemiology of Pediatric Traumatic Brain Injury and Hypothalamic-Pituitary Disorders in Arizona. *Front Neurol*. 2020 Jan 22;10:1410. doi: 10.3389/fneur.2019.01410. eCollection 2019. PubMed PMID: 32038466; PubMed Central PMCID: PMC6988738.

Palmqvist S, Janelidze S, Quiroz YT, Lopera F, Stomrud E, Su Y, Chen Y, Serrano GE, Leuzy A, Mattsson N, Strandberg O, Smith R, Villegas A, Sepulveda-Falla D, Chai X, Proctor NK, Zetterberg H, Beach TG, Blennow K, Dage JL, Reiman EM, Hansson O. Diagnostic accuracy of plasma phospho-tau217 for Alzheimer's disease (submitted).

Paramasivan S, Bassiouni A, Shiffer A, Dillon MR, Cope EK, Cooksley C, Ramezanzpour M, Moraitis S, Ali MJ, Bleier B, Callejas C, Cornet ME, Douglas RG, Dutra D, Georgalas C, Harvey RJ, Hwang PH, Luong AU, Schlosser RJ, Tantilipikorn P, Tewfik MA, Vreugde S, Wormald PJ,

Caporaso JG, Psaltis AJ. The international sinonasal microbiome study: A multicentre, multinational characterization of sinonasal bacterial ecology. *Allergy*. 2020 Mar 13. doi: 10.1111/all.14276. [Epub ahead of print] PubMed PMID: 32167574.

Piras IS, Bleul C, Talboom JS, De Both MD, Schrauwen I, Halliday G, Myers AJ, Serrano GE, Beach TG, Huentelman MJ. ESHRD: deconvolution of brain homogenate RNA expression data to identify cell-type-specific alterations in Alzheimer's disease. *Aging (Albany NY)*. 2020 Mar 2;12(5):4124-4162. doi: 10.18632/aging.102840. Epub 2020 Mar 2. PubMed PMID: 32125278; PubMed Central PMCID: PMC7093163.

Piras, Ignazio S. et al "Transcriptional profiling of Multiple System Atrophy cerebellar tissue highlights differences between the parkinsonian and cerebellar sub-types of the disease." *bioRxiv* (2020): 2020.02.11.944306. Web. 06 April. 2020 (submitted)

Pluvinage JV, Wyss-Coray T. Systemic factors as mediators of brain homeostasis, ageing and neurodegeneration. (PMID 31913356) *Nat Rev Neurosci*. 2020 Feb;21(2):93-102. doi: 10.1038/s41583-019-0255-9. Epub 2020 Jan 8. (indirectly)

Polsinelli AJ, Moseley SA, Grilli MD, Glisky EL, Mehl MR. Natural, Everyday Language Use Provides a Window into the Integrity of Older Adults' Executive Functioning. *J Gerontol B Psychol Sci Soc Sci*. 2020 Apr 20. pii: gbaa055. doi: 10.1093/geronb/gbaa055. [Epub ahead of print] PubMed PMID: 32310293.

Polsinelli A, Rentscher K, Glisky E, Moseley S, Mehl M. (2020). Interpersonal Focus in the Emotional Autobiographical Memories of Older and Younger Adults. *GeroPsych*. 33. 3-14. 10.1024/1662-9647/a000220.

Qi G, Mi Y, and Yin F. (2020) Cellular specificity and inter-cellular coordination in the brain bioenergetic system: Implications for aging and neurodegeneration. *Front Physiol.*, 10:1531. doi: 10.3389/fphys.2019.01531.

Quiroz, Y, Zetterberg H, Reiman E, Chen Y, Su Y, García G, Villegas A, Sepulveda-Falla D, Villada M, Arboleda-Velasquez J, Fox-Fuller J, Guzmán-Vélez E, Vila-Castelar C, Gordon BA, Schultz S, Protas H, Ghisays V, Giraldo M, Tirado V, Baena A, Munoz C, Rios-Romenets S, Tariot P, Blennow K, Lopera F. (2020). Plasma neurofilament light measurements in more than 2,100 presenilin-1 E280A mutation carriers and non-carriers from the world's largest autosomal dominant Alzheimer's disease kindred. *The Lancet Neurology*. (Accepted)

Raichlen DA and Alexander GE (in press) Why your brain needs exercise: Key transitions in the evolutionary history of humans may have linked body and mind in ways that we can exploit to slow brain aging. *Scientific American*.

Raichlen DA, Bharadwaj PK, Nguyen LA, Franchetti MK, Zigman EK, Solorio AR, Alexander GE. Effects of simultaneous cognitive and aerobic exercise training on dual-task walking performance in healthy older adults: results from a pilot randomized controlled trial. *BMC Geriatr*. 2020 Mar 2;20(1):83. doi: 10.1186/s12877-020-1484-5. PubMed PMID: 32122325; PubMed Central PMCID: PMC7052975.

Ratiu I, Azuma T. 2020. Assessment of executive function in bilingual adults with history of mild traumatic brain injury. *Brain Impairment*. In press.

Reiman EM, Arboleda-Velasquez JF, Quiroz YT, Huentelman MJ, Beach TG, Caselli RJ, Chen Y, Su Y, Myers AJ, Hardy J, Paul Vonsattel J, Younkin SG, Bennett DA, De Jager PL, Larson EB, Crane PK, Keene CD, Kamboh MI, Kofler JK, Duque L, Gilbert JR, Gwirtsman HE, Buxbaum JD, Dickson DW, Frosch MP, Ghetti BF, Lunetta KL, Wang LS, Hyman BT, Kukull WA, Foroud T, Haines JL, Mayeux RP, Pericak-Vance MA, Schneider JA, Trojanowski JQ, Farrer LA, Schellenberg GD, Beecham GW, Montine TJ, Jun GR; Alzheimer's Disease Genetics Consortium. Exceptionally low likelihood of Alzheimer's dementia in APOE2 homozygotes from a 5,000-person neuropathological study. *Nat Commun.* 2020 Feb 3;11(1):667. doi: 10.1038/s41467-019-14279-8. PubMed PMID: 32015339.

Rios-Romenets S, Lopera F, Sink KM, Hu N, Lian Q, Guthrie H, Smith J, Cho W, Mackey H, Langbaum JB, Thomas RG, Giraldo-Chica M, Tobon C, Acosta-Baena N, Munoz C, Opsina P, Tirado V, Henao E, Boganegra Y, Chen K, Su Y, Goradia D, Thiyyagura P, VanGilder PS, Luo J, Ghisays V, Lee W, Malek-Ahmadi MH, Protas HD, Chen Y, Quiroz YT, Reiman EM, Tariot PN, API ADAD Trial Group. Baseline demographic, clinical and cognitive characteristics of the Alzheimer's Prevention Initiative (API) Autosomal-Dominant Alzheimer's Disease Colombia Trial. *Alzheimer's & Dementia*, in-press.

Roberts-Wolfe DJ, Heinsbroek JA, Spencer SM, Bobadilla AC, Smith ACW, Gipson CD, Kalivas PW. Transient synaptic potentiation in nucleus accumbens shell during refraining from cocaine seeking. *Addict Biol.* 2020 May;25(3):e12759. doi: 10.1111/adb.12759. Epub 2019 May 6. PubMed PMID: 31062493.

Sabat J, Bock D, Hsu CH, Tan TW, Weinkauff C, Trouard T, Perez-Carrillo GG, Zhou W. Risk factors associated with microembolization after carotid intervention. *J Vasc Surg.* 2020 May;71(5):1572-1578. doi: 10.1016/j.jvs.2019.06.202. Epub 2019 Sep 5. PubMed PMID: 31493967; PubMed Central PMCID: PMC7054135.

Schechter M, Grigoletto J, Abd-Elhadi S, Glickstein H, Friedman A, Serrano GE, Beach TG, Sharon R. A role for  $\alpha$ -Synuclein in axon growth and its implications in corticostriatal glutamatergic plasticity in Parkinson's disease. *Mol Neurodegener.* 2020 Mar 30;15(1):24. doi: 10.1186/s13024-020-00370-y. PubMed PMID: 32228705; PubMed Central PMCID: PMC7104492.

Sekar, Shobana et al "ACValidator: a novel assembly-based approach for in silico validation of circular RNAs." *bioRxiv* (2019): 556597. Web. 06 April. 2020 (submitted)

Serrano GE, Shprecher D, Callan M, Cutler B, Glass M, Zhang N, Walker J, Intorcchia A, Adler CH, Shill HA, Driver-Dunckley E, Mehta SH, Belden CM, Zamrini E, Sue LI, Vargas D, Beach TG. Cardiac sympathetic denervation and synucleinopathy in Alzheimer's disease with brain Lewy body disease. *Brain Commun.* 2020;2(1):fcaa004. doi: 10.1093/braincomms/fcaa004. Epub 2020 Jan 28. PubMed PMID: 32064463.

Shang Y, Mishra A, Wang T, Wang Y, Desai M, Chen S, Mao Z, Do L, Bernstein AS, Trouard TP, and Brinton RD (2020) Evidence in support of chromosomal sex influencing plasma based metabolome vs APOE genotype influencing brain metabolome profile in humanized APOE male and female mice. *PLoS ONE*, doi: 10.1371/journal.pone.0225392. eCollection.

Shen X, Ma S, Vemuri P, Simon G; Alzheimer's Disease Neuroimaging Initiative. Challenges and Opportunities with Causal Discovery Algorithms: Application to Alzheimer's Pathophysiology. *Sci Rep.* 2020 Feb 19;10(1):2975. doi: 10.1038/s41598-020-59669-x. PMID: 32076020

Shprecher DR, Adler CH, Zhang N, Shill HA, Belden CM, Driver-Dunckley E, Mehta SH, Davis KJ, Sue LI, Zamrini E, Beach TG. Do Parkinson disease subject and caregiver-reported Epworth sleepiness scale responses correlate? *Clin Neurol Neurosurg*. 2020 Feb 6;192:105728. doi: 10.1016/j.clineuro.2020.105728. [Epub ahead of print] PubMed PMID: 32058206.

Shprecher DR, Serrano GE, Zhang N, Intorcica A, Davis KJ, Glass M, Curry J, Walker J, Cutler B, Callan M, Garcia A, Sue LI, Beach TG. Prevalence of REM sleep behavior disorder in Sun City, Arizona. *Heliyon*. 2020 Jan 9;6(1):e03140. doi: 10.1016/j.heliyon.2019.e03140. eCollection 2020 Jan. PubMed PMID: 31938745.

Srinivasan, Karpagam et al "Alzheimer's patient brain myeloid cells exhibit enhanced aging and unique transcriptional activation." *bioRxiv* (2019): 610345. Web. 06 April. 2020. (submitted)

Stokes AM, Semmineh NB, Nespodzany A, Bell LC, Quarles CC. "Systematic Assessment of Multi-Echo Dynamic Susceptibility Contrast (DSC) MRI using a Digital Reference Object (DRO)" *Magnetic Resonance in Medicine* 83(1):109-123 (2020).

Talmage, C., Coon, D. W., Dugger, B. N., Knopf, R. C., O'Connor, K. A., & Schofield, S. A. (2020). Social leisure activity, physical activity, and valuation of life: Findings from a Longevity Study. *Activities, Adaptation & Aging*, 44, 61-84.

Tang YY, Fan Y, Lu Q, Tan LH, Tang R, Kaplan RM, Pinho MC, Thomas BP, Chen K, Friston KJ, Reiman EM. Long-Term Physical Exercise and Mindfulness Practice in an Aging Population. *Front Psychol*. 2020 Apr 2;11:358. doi: 10.3389/fpsyg.2020.00358. eCollection 2020. PubMed PMID: 32300317; PubMed Central PMCID: PMC7142262.

Tu Y, Mi L, Zhang W, Zhang H, Zhang J, Fan Y, Goradia D, Chen K, Caselli RJ, Reiman EM, Gu X, Wang Y; Alzheimer's Disease Neuroimaging Initiative. Computing Univariate Neurodegenerative Biomarkers with Volumetric Optimal Transportation: A Pilot Study. *Neuroinformatics*. 2020 Apr 6. doi: 10.1007/s12021-020-09459-7. [Epub ahead of print] PubMed PMID: 32253701.

Valdez MA, Fernandez E, Matsunaga T, Erickson RP, and Trouard, TP (2020) Distribution and diffusion of macromolecule delivery to the brain via focused ultrasound using magnetic resonance and multispectral fluorescence Imaging. *Ultrasound in Medicine & Biology*, 46:122-136.

Walker DG, Tang TM, Mendsaikhan A, Tooyama I, Serrano GE, Sue LI, Beach TG, Lue LF. Patterns of Expression of Purinergic Receptor P2RY12, a Putative Marker for Non-Activated Microglia, in Aged and Alzheimer's Disease Brains. *Int J Mol Sci*. 2020 Jan 20;21(2). pii: E678. doi: 10.3390/ijms21020678. PubMed PMID: 31968618.

Walpola IC, Muller AJ, Hall JM, Andrews-Hanna JR, Irish M, Lewis SJG, Shine JM, O'Callaghan C. Mind-wandering in Parkinson's disease hallucinations reflects primary visual and default network coupling. *Cortex*. 2020 Apr;125:233-245. doi: 10.1016/j.cortex.2019.12.023. Epub 2020 Jan 22. PubMed PMID: 32058090.

Wang Y, Hernandez G, Mack WJ, Schneider LS, Yin F, Brinton RD. Retrospective analysis of phytoSERM for management of menopause-associated vasomotor symptoms and cognitive decline: a pilot study on pharmacogenomic effects of mitochondrial haplogroup and APOE genotype on therapeutic efficacy. *Menopause*. 2020 Jan;27(1):57-65. doi:

10.1097/GME.0000000000001418. PubMed PMID: 31567873; PubMed Central PMCID: PMC7100617.

Wang Z, Yu K, Tan H, Wu Z, Cho JH, Han X, Sun H, Beach TG, Peng J. 27-Plex Tandem Mass Tag Mass Spectrometry for Profiling Brain Proteome in Alzheimer's Disease. *Anal Chem*. 2020 May 7. doi: 10.1021/acs.analchem.0c00655. [Epub ahead of print] PubMed PMID: 32343560.

Wang Y, Mishra A, and Brinton RD (2020) Transitions in metabolic and immune systems from pre-menopause to post-menopause: implications for age-associated neurodegenerative diseases. *F1000 Faculty Reviews*, doi: 10.12688/f1000research.21599.1. eCollection 2020.

Wijasa TS, Sylvester M, Brocke-Ahmadinejad N, Schwartz S, Santarelli F, Gieselmann V, Klockgether T, Brosseron F, Heneka MT. Quantitative proteomics of synaptosome S-nitrosylation in Alzheimer's disease. *J Neurochem*. 2020 Mar;152(6):710-726. doi: 10.1111/jnc.14870. Epub 2019 Oct 21. PMID: 31520481

Xavier JB, Young VB, Skufca J, Ginty F, Testerman T, Pearson AT, Macklin P, Mitchell A, Shmulevich I, Xie L, Caporaso JG, Crandall KA, Simone NL, Godoy-Vitorino F, Griffin TJ, Whiteson KL, Gustafson HH, Slade DJ, Schmidt TM, Walther-Antonio MRS, Korem T, Webb-Robertson BM, Styczynski MP, Johnson WE, Jobin C, Ridlon JM, Koh AY, Yu M, Kelly L, Wargo JA. The Cancer Microbiome: Distinguishing Direct and Indirect Effects Requires a Systemic View. *Trends Cancer*. 2020 Mar;6(3):192-204. doi: 10.1016/j.trecan.2020.01.004. Epub 2020 Feb 7. Review. PubMed PMID: 32101723; PubMed Central PMCID: PMC7098063.

Yan T, Liang J, Gao J, Wang L, Fujioka H; Alzheimer Disease Neuroimaging Initiative, Zhu X, Wan FAM222A encodes a protein which accumulates in plaques in Alzheimer's disease. *Nat Commun*. 2020 Jan 21;11(1):411. doi: 10.1038/s41467-019-13962-0. PMID: 31964863

## Current and Pending Grants

### Current Grants

Heather Bimonte-Nelson (PI)  
R01 AG028084 (Bimonte-Nelson) 9/01/07 - 8/31/23  
National Institute on Aging \$1,828,473 Total Costs  
Variations in hormones during menopause: effects on cognitive and brain aging

Heather Bimonte-Nelson (PI)  
R01 Grant, Diversity Supplement 5/17-12/19  
National Institute on Aging \$50,000 Total Costs  
Variations in hormone therapy: effects on cognition and markers of brain aging

Heather Bimonte-Nelson (Core Co-Leader)  
2P30AG19610 (Reiman) 7/1/01-6/30/21  
NIH/NIA  
Arizona Alzheimer's Disease Core Center – Research Education Component

Heather Bimonte-Nelson (Co-I)  
R01 NS097537 (Newbern) 7/1/2016 – 5/31/2021  
NIH-HHS (NINDS) \$1,731,322 Total Costs  
Functions of ERK/MAPK Signaling in GABAergic Circuit Development

Heather Bimonte-Nelson (Co-I)  
R01 DA043172 (Foster Olive) 9/26/2017 – 8/31/2022  
NIH \$1,875,000 Total Costs  
Characterization and reversal of neurocognitive dysfunction produced by long-term synthetic cathinone use

Heather Bimonte-Nelson (Associate Director)  
T32AG044402 (Barnes) 5/01/2016 – 4/30/2021  
NIH/NIA \$1,237,680 Total Costs  
Postdoctoral Training, Neurobiology of Aging and Alzheimer's Disease

Heather Bimonte-Nelson (Co-I)  
CTR040636 (Coon, David) 7/1/2019-12/31/2020  
Arizona Alzheimer's Consortium \$56,667  
Arizona Alzheimer's Consortium (AAC) FY20

David Brafman (Project PI)  
Arizona Alzheimer's Disease Consortium (PI: Brafman) 07/01/2019-06/30/2020  
FY 2020 AADC \$60,000 Total Costs  
Using hiPSC-models to establish a causative link between traumatic brain injury (TBI), neuroinflammation, and Alzheimer's disease

David Brafman (PI)  
R21 AG063358 (PI: Brafman) 04/01/2019-03/31/2021  
NIH/NIA \$434,336 Total Costs  
A Pluripotent Stem Cell-Based Model to Investigate the Mechanisms of TBI-Induced

David Brafman (PI) T0138 (PI: Brafman) Department of Defense Adaptable multi-modality nanoprobes for non-invasive real-time monitoring of engineered cells and tissues	10/26/2018-10/25/2020 \$1,250,878 Total Costs
David Brafman (PI) Arizona Board of Regents (PI: Brafman) Statewide Collaborative Regenerative Medicine Research and Training Facility	07/1/2018-12/31/2019 \$400,000 Total Costs
David Brafman (PI) T0042-C (PI: Brafman) Department of Defense Total Costs: Biomanufacturing of Cells in the Neuroectoderm Fate Space	01/01/2018-03/31/2021 \$1,387,138 Total Costs
David Brafman (PI) R21 AG056706 (PI: Brafman) NIH/NIA Generation and characterization of isogenic hiPSC lines with various APOE genotypes	09/15/2017-04/30/2020 \$409,034 Total Costs
David Brafman (PI) GM121698 (PI: Brafman) NIH-NIGMS Investigating the mechanisms of a multi-state model of Wnt signaling	04/01/2017-03/31/2022 \$1,518,984 Total Costs
David Brafman (PI) ADHS16-162401 (PI: Brafman) ABRC Using human induced pluripotent stem cells to investigate the contribution of risk variants and aging to the onset and progression of Alzheimer's disease.	04/01/2017-03/31/2021 \$225,000 Total Costs
David Coon (PI) R01AG049895 (Coon) NIH-NIA EPIC: A group-based intervention for Early-stage AD Dyads in Diverse Communities	05/15/16 – 04/30/22 \$1,486,917
David Coon (OR Core Leader) P30 AG019610 (Reiman) NIA Arizona Alzheimer's Disease Center Outreach and Recruitment Core	07/01/2016 - 6/30/2021 \$57,216
David Coon (Co-PI) AWD3052 (Coon) Arizona Alzheimer's Disease Consortium Psychosocial Intervention Development for Those Living alone with Mild Cognitive Impairment	07/01/18 – 06/30/19 \$80,000
David Coon (PI) 90ALGG0019-01-00 (Coon) HHS-ACL ADI-SSS Arizona's Dementia-Capable System Enhancement	09/01/17 – 08/31/20 \$808,150

David Coon (Co-PI) AWD30399 (UofAz) Dignity Health-St. Joseph's Hospital: BNI Parkinson's Partners in Care: Focus Group and Pilot	01/01/19 – 9/30/20 \$181,816
David Coon (PI) AWD33102 (Coon) Phoenix Symphony Music and Memory II	08/01/18 – 07/31/20 \$100,000
David Coon (PI) AWD34006 (Coon) Valley of the Sun United Way VSUW / HOPE Promotores: Cartwright, Alhambra, Central City, Guadalupe	03/15/19 – 06/30/20 \$100,000
David Coon (PI) CTR040636 (Coon) Arizona Alzheimer's Consortium Piloting an Evidence-based Intervention for Those Living alone with Mild Cognitive Impairment	07/01/19 – 06/30/20 \$640,833
David Coon (Co-I) FP21036 (Underiner) National Endowment for the Arts (NEA) Creative Health Collaborations Hub	03/01/20 – 02/28/22 \$63,881
Cassandra Gipson-Reichardt (PI) R21 DA044479 National Institute on Drug Abuse. Cholinergic modulation of glutamatergic signaling in nicotine addiction and relapse	07/18-06/20
Cassandra Gipson-Reichardt (PI) R21 DA044479-S1 Diversity Supplement: from National Institute on Drug Abuse. Cholinergic modulation of glutamatergic signaling in nicotine addiction and relapse	01/19-06/20
Cassandra Gipson-Reichardt (PI) R03 DA045881 from National Institute on Drug Abuse. Glutamatergic mechanisms underlying nicotine addiction and relapse following nicotine reduction	09/18-08/20
Cassandra Gipson-Reichardt (PI) R01 DA046526 from National Institute on Drug Abuse. Neuroinflammatory and glutamatergic mechanisms of nicotine seeking	09/19-08/24
Cassandra Gipson-Reichardt (Co-I) R01 DA043172 (Foster Olive) from National Institute on Drug Abuse. Characterization and reversal of neurocognitive dysfunction produced by long-term synthetic cathinone use	09/18-7/22

Cassandra Gipson-Reichardt (Co-I) R01 DA043172 (Foster Olive) from National Institute on Drug Abuse. Contributions of Glial Glutamate Transport and NMDA Receptors in Nicotine Relapse	01/16-12/19
Cassandra Gipson-Reichardt (Subcontract PI) R41 Grant (Zaczek) from National Institute on Drug Abuse. Preclinical assessment of the GluN2B inhibitor clinical candidate NP10679 as a medication to prevent opiate abuse	7/18-6/20
Diego Mastroeni (PI) 17-0473-ASU (Mastroeni) Arizona Veterans Research and Education Foundation Probing the Mechanistic Role of Vascular Dysfunction and Vascular Inflammation in TBI-Mediated Cognitive Dysfunction	8/1/2017-7/31/2020 \$27,126
Diego Mastroeni (PI) AARGD-17-529197 (Mastroeni) Arizona State University Foundation (ASUF) Gender Effects on Identified Cell Population in Alzheimer's Disease	3/1/2018-2/28/2021 \$136,364
Diego Mastroeni (Co-I) AGR 08/22/18 (Readhead) Banner Alzheimer's Institute A Public Resource of RNA Sequencing Data from Different Human Brain Cells and Regions, Associated Whole Genome Sequencing, Longitudinal Clinical and Neuropathological Data, and Cell-Specific Multi-Scale Networks in the Alzheimer's and Aging Brain	7/1/2017-6/30/2021 \$289,492
Diego Mastroeni (Project PI) CTR040636 (Coon) Arizona Alzheimer's Consortium FY20 Are PSEN1 E280A mutation carriers an accelerated form of Dementia with Lewy body, not Alzheimer's disease?	7/1/2019-12/31/2020 \$640,833
Benjamin Readhead (PI) AGR 08/22/18 (Readhead) Banner Alzheimer's Institute A Public Resource of RNA Sequencing Data from Different Human Brain Cells and Regions, Associated Whole Genome Sequencing, Longitudinal Clinical and Neuropathological Data, and Cell-Specific Multi-Scale Networks in the Alzheimer's and Aging Brain	7/1/2017-6/30/2021 \$289,492
Benjamin Readhead (PI) U01AG061835 (Readhead) HHS: National Institutes of Health (NIH) Identification of the genetic and transcriptomic networks of cognitive and neuropathological resilience to Alzheimer's disease associated viruses	9/1/2018-8/31/2023 \$1,998,866
Benjamin Readhead (PI) R21AG063068 (Readhead) HHS: National Institutes of Health (NIH)	4/1/2019-1/31/2021 \$323,397

Investigation of chromosomally integrated Human Herpesvirus 6 as a risk factor for Alzheimer's disease

Benjamin Readhead (Co-I)  
R01 AG062500 (Velazquez, Ramon) 4/15/2019-2/29/2024  
HHS: National Institutes of Health (NIH) \$347,994  
S6K1 as a novel link between aging and Alzheimer's disease

Benjamin Readhead (PI)  
AGR 05/7/19 (Readhead) 5/10/2019-5/10/2020  
Global Lyme Alliance \$67,501  
An interesting necroptosis angle: tick-borne disease and AD

Benjamin Readhead (Project PI)  
CTR040636 (Coon) 7/1/2019-12/31/2020  
Arizona Alzheimer's Consortium (AAC) FY20 \$640,833  
Multiomic modelling of microbe-host interactions in the brain affected by late onset Alzheimer's disease

Benjamin Readhead (PI)  
2019-26 (Readhead) 9/1/2019-9/1/2020  
Arizona State University Foundation (ASUF) \$500,000  
Characterizing the microbiome of preclinical and early stage Alzheimer's disease and additional neurodegenerative diseases.

Benjamin Readhead (PI)  
UWSC11621 (Readhead) 9/1/2019-5/31/2024  
University of Washington \$11,609  
Modulation of Alzheimer's disease by Herpes simplex virus infection

Sarah Stabenfeldt (PI)  
1454282 (Stabenfeldt) 04/01/15-03/31/21  
NSF-CBET  
CAREER: Elucidation and modulation of chemotactic signaling after brain injury

Sarah Stabenfeldt (PI)  
ADHS18-198843 (Stabenfeldt) 04/01/18-03/31/21  
AZ DEPT OF HEALTH SERVICES  
Regenerative rehabilitation for traumatic brain injury

Sarah Stabenfeldt (MPI)  
R21 NS107985 (Stabenfeldt/Sirianni) 06/01/18-05/31/21  
HHS-NIH-NINDS  
Nanotherapeutics to alleviate neuroinflammation after TBI

Sarah Stabenfeldt (MPI)  
R21 AG063358 (Brafman/Stabenfeldt) 04/01/19-03/31/21  
HHS-NIH-NIA  
A Pluripotent Stem Cell-Based Model to Investigate the Mechanisms of TBI-Induced AD

Yalin Wang (PI) RF1AG051710 University of Michigan Multi-Source Sparse Learning to Identify MCI and Predict Decline	6/1/2016-5/31/2020 \$536,000
Yalin Wang (PI) R01EB025032 Children's Hospital Los Angeles Predicting the early childhood outcomes of preterm brain shape abnormalities	9/22/2017 – 6/30/2021 \$68,080
Yalin Wang (Project PI) Arizona Alzheimer's Consortium (Coon) State of Arizona FY20 Arizona Alzheimer's Disease Consortium	7/1/2018-6/30/2019 \$30,000
Yalin Wang (PI) R01EB025032-03S1 Children's Hospital Los Angeles Predicting the Early Childhood Outcomes of Preterm Brain Shape Abnormalities	9/21/2019-6/30/2020 \$42,365
Melissa Wilson (PI) NIH MIRA Population dynamics and medical consequences of sex chromosome evolution	09/2017 – 08/2022 \$1,650,000
Melissa Wilson (Co-I) Breast Cancer Research Foundation (Anderson) Targeting breast cancer tumor antigens for immunotherapy PI: Anderson; Co-I: Wilson Sayres (5%; \$12,500); Co-I: Borges	10/2019 – 09/2020 \$250,000
Melissa Wilson (Co-I) AZ Alzheimer's Disease Consortium	07/2019 – 06/2020 \$300,000
Eric Reiman (PI) 5 P30 AG19610 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center	9/30/2001-6/30/2021 \$1,628,650 Annual DC
Eric Reiman (PI) 5 R01 AG031581 (Reiman) NIH/NIA Brain Imaging, APOE, & the Preclinical Course of Alzheimer's Disease	5/01/2008-3/31/2021 \$1,350,755 Annual DC
Eric Reiman (PI) 5R01AG055444-02 (Reiman/Tariot/Lopera) NIH/NIA Alzheimer's Prevention Initiative ADAD Colombia Trial	4/1/2017-3/31/2022 \$1,582,894 Annual DC
Eric Reiman (Co-I) SAGA-17-415540 (Rasgon) Alzheimer's Association/Leland Stanford Jr University Sex Specific Interactions of Modifiable and non-Modifiable Risk Factor of AD	12/01/2017-11/30/2019 \$36,263 Annual DC

Eric Reiman (PI) U01NS093334-04 (Cummings/Reiman/Shenton/Stern) NIH/NINDS Boston University via Mayo Clinic Arizona Chronic Traumatic Encephalopathy: Detection, Diagnosis, Course and Risk Factors	12/15/2015-11/30/2022 \$1,969,873 Annual DC
Eric Reiman (PI) Alzheimer's Association/GHR/FBRI (Reiman/Tariot/Langbaum) Alzheimer's Prevention Initiative APOE4 Trial	1/1/2016-12/31/2020 \$10,000,000 TC
Eric Reiman (PI) 1OT2OD026549-01 (Reiman) NIH/University of Arizona University of Arizona-Banner Health All of Us Research Program	04/01/2018-3/31/2023 \$958,240. Annual DC
Eric Reiman (PI) NOMIS Foundation (Reiman) NOMIS Foundation via Banner Alzheimer's Foundation A Public Resource of RNA Sequencing Data from Different Human Brain Cells and Regions, Associated Whole Genome Sequencing, Longitudinal Clinical and Neuropathological Data, and Cell-Specific Multi-Scale Networks in the Alzheimer's and Aging Brain	9/1/2007-8/31/2021 \$1,240,381 Annual DC
Eric Reiman (Consortium PI) P01AG052350-03 (Zlokovic/Toga) NIH via USC Vascular Contributions to Dementia and Genetic Risk Factors for Alzheimer's Disease	9/30/2016 – 5/31/2021 \$74,670 Annual DC
Eric Reiman (Consortium PI) 5R01AG054671-02 (Quiroz) NIA/NIA via Harvard University Relationship between tau pathology and cognitive impairment in autosomal dominant Alzheimer's disease	09/01/2017-5/31/2022 \$33,040 Annual DC
Eric Reiman (Collaborator) RF1AG054617 (Pa) NIH via USC Gender and APOE4 effects on brain morphometry, cognition, and clinical progression to Alzheimer's Disease	09/01/2017 - 08/31/2022 \$11,318 Annual DC
Eric Reiman (Co-I) U19AG024904 (Weiner) NIA/Northern California Institute Res & Educ. Alzheimer's Disease Neuroimaging Initiative	09/30/2017-7/31/2021 \$85,000 Annual DC
Eric Reiman (PI) 1R01AG058468 (Reiman/Tariot/Langbaum/Aisen/ Sperling/Johnson) NIH/NIA API / A4 Alzheimer's Prevention Trial	9/1/2018-11/30/24 \$4,085,152 Annual DC
Eric Reiman (PI) P30 AG19610-19S1A1 (Reiman)	9/15/2018 -6/30/2021

NIH/NIA Arizona Alzheimer's Disease Core Center – Brain Imaging and Fluid Biomarker Core	\$1,242,362 Annual DC
Eric Reiman (Consortium PI) U54MD000507 (Manson/Buchwald) University of Colorado Denver/NIH/NIMHH American Indian and Alaska Native Health Disparities	9/22/2017-4/30/2022 \$50,000 Annual DC
Eric Reiman (Co-I) U01AG016976 (Kukull) NIA/University of Washington National Alzheimer's Coordinating Center	07/01/2014-6/30/2020 \$159,900
Yi Su (Co-I) R01 AG031581 (Reiman) NIH/NIA Brain Imaging, APOE, & the Preclinical Course of Alzheimer's Disease	05/01/08-03/31/20 \$1,350,755 Annual DC
Yi Su (Site Co-I) U54MD000507 (Manson/Nelson) University of Colorado Denver/NIH/NIMHH American Indian and Alaska Native Health Disparities	9/22/2017-4/30/2020 \$50,000 Annual DC
Yi Su (Co-I) ASU Grant ID PG08347(Su) Arizona State University	7/1/2019-6/30/2020 \$137,334 Annual DC
Yi Su (Co-I) BNI Grant ID BAI33132 (Stokes) Dignity Health Multi-Scale MRI Assessment of Neurovascular Factors Associated with AD	7/1/2019-6/30/2020 \$34,376 Annual DC
Yi Su (PI) BrightFocus ADR A2017272S (Su) BrightFocus Foundation Blood Brain Barrier and Metabolism in Aging and Alzheimer Disease	7/1/2017-6/30/2020 \$300,000 TC
Yi Su (PI) AARG17532945 (Su) Alzheimer's Association Amyloid PET as a biomarker for white matter integrity in Alzheimer disease	10/1/2017-9/30/2020 \$150,000 TC
Yi Su (Project PI) Arizona Alzheimer's Research Consortium (Reiman) State of Arizona Advanced Image Analysis Techniques for the Detection and Tracking of Alzheimer's disease and its prevention	7/1/2018-6/30/2020 \$150,000 Annual DC
Yi Su (Project PI) Arizona Alzheimer's Research Consortium (Reiman)	7/1/2018-6/30/2020

State of Arizona Statistical and Neuroimaging Core Resources Serving the Consortium members for the Alzheimer's disease and prevention related studies	\$50,000 Annual DC
Yi Su (Project PI) 5R01AG055444-02 (Reiman/Tariot/Lopera) NIH/NIA Alzheimer's Prevention Initiative ADAD Colombia Trial	4/15/2018-3/31/2023 \$1,582,894 Annual DC
Yi Su (Co-I) U19AG024904 (Weiner) NIA/Northern California Institute Res & Educ. Alzheimer's Disease Neuroimaging Initiative	9/30/2017-7/31/2021 \$43,809 Annual DC
Yi Su (Consortium PI) SAGA-17-415540 (Rasgon) Alzheimer's Association/Leland Stanford Jr. University Sex Specific Interactions of Modifiable and Non-Modifiable Risk Factor of AD	12/1/2017-11/30/19 \$36,263 Annual DC
Yi Su (Co-I) 1R01AG058468 (Reiman/Tariot/Langbaum/Aisen/Sperling/Johnson) NIH/NIA API/ A4 Alzheimer's Prevention Trial	9/1/2018-8/31/2023 \$4,085,152 Annual DC
Yi Su (PI) R21EB024366 (Wang/Goyal/Su) NIH/NBIB via Cornell University Feasibility of challenge-free QSM based quantitative mapping of cerebral metabolic rate of oxygen	04/01/17-01/31/20 \$17,000 Annual DC
Kewei Chen (Co-I/Core Leader) P30 AG019610 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center	07/01/16-6/30/21 \$1,628,650 Annual DC
Kewei Chen (Co-I) 1R01AG055444-01 (Reiman/Tariot/Lopera) NIH/NIA Alzheimer's Prevention Initiative ADAD Colombia Trial	04/1/17-3/31/22 \$1,582,894 Annual DC
Kewei Chen (Co-I) R01 AG031581 (Reiman) NIH/NIA Brain Imaging, APOE, & the Preclinical Course of Alzheimer's Disease	05/01/08-03/31/20 \$1,350,755 Annual DC
Michael Malek-Ahmadi (Consortium PI) 1RF1AG057547-01 (Kantarci/Gleason) NIA/NIA via Mayo Clinic Prevention of Alzheimer's disease in women: risks and benefits of hormone therapy	09/15/07-06/30/22

Michael Malek-Ahmadi (Co-I/Core Leader) P01AG014449 (Mufson) NIH/NIA via Dignity Health Neurobiology of Mild Cognitive Impairment in the Elderly	03/15/19 – 01/31/21
Michael Malek-Ahmadi (Co-I) Arizona Alzheimer's Research Consortium (Reiman) State of Arizona Department of Human Services Advanced Image Analysis Techniques for the Detection and Tracking of Alzheimer's disease and its prevention	7/1/19-6/30/20 \$150,000 Annual DC
Michael Malek-Ahmadi (Co-I) Arizona Alzheimer's Research Consortium (Reiman) State of Arizona Department of Human Services Statistical and Neuroimaging Core Resources Serving the Consortium members for the Alzheimer's disease and prevention related studies	7/1/19-6/30/20 \$50,000 Annual DC
Jessica Langbaum (Co-Investigator) 5R01AG055444 (Reiman/Tariot/Lopera) NIH/NIA Alzheimer's Prevention Initiative ADAD Colombia Trial	04/01/17-03/31/22 \$1,582,894 Annual DC
Jessica Langbaum (Project PI) Arizona Alzheimer's Research Consortium (Reiman) AAC via State of Arizona DHS Arizona Alzheimer's Registry	07/01/11-06/30/19 \$12,149 Annual DC
Jessica Langbaum (Co-PI) Alzheimer's Association/GHR/FBRI (Reiman/Tariot/Langbaum) Alzheimer's Prevention Initiative APOE4 Trial	01/01/16-12/31/20 \$10,000,000 Total Costs
Jessica Langbaum (Co-PI) 1R01AG058468 (Reiman/Tariot/Langbaum/ Aisen/Sperling/Johnson) NIA/NIA API / A4 Alzheimer's Prevention Trial	09/01/18-11/30/24 \$4,085,182
Don Saner (Project PI) Arizona Alzheimer's Research Consortium (Reiman) State of Arizona Enhancements to a Centralized Data Management System For the Arizona Alzheimer's Disease Core Center (ADCC), Brain and Body Donation Program (BBDP), and Apolipoprotein E4 (APOE4) Gene Dose Program	07/01/19-06/30/20 \$100,000 Annual DC
Don Saner (Core Leader) 5P30AG019610 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center	09/30/01-06/30/21 \$1,628,650 Annual DC
Don Saner (Data Science Sr. Director) 3OT2OD026549 (Ojo/Reiman/Theodorou)	04/01/18-03/31/23 \$958,240

NIH/NIA via University of Arizona  
University of Arizona-Banner Health All of Us Research Program

Don Saner (Co-I) E01AG058468 (Reiman/Aisen/Johnson/ Langbaum/Sperling/Tariot) NIH/NIA API / A4 Alzheimer's Prevention Trial	09/01/18-11/30/24 \$32,005,950 TC
Danielle Goldfarb (Project PI) Arizona Alzheimer's Research Consortium (Reiman) AAC via State of Arizona DHS Advancing the Use of Lumbar Puncture in Alzheimer's Disease and Related Disorders Research	07/01/19-06/30/20 \$25,000 Annual DC
David Weidman (Site PI) U24 AG057437 (Aisen) NIH/NIA via USC (ATRI) Alzheimer's Clinical Trial Consortium	12/02/17-11/30/22 \$896,667 Total Project Costs
David Weidman (Site PI) R01 AG053798 (Aisen) NIH/NIA via USC (ATRI) Global Alzheimer's Platform Trial-Ready Cohort for Preclinical/Prodromal Alzheimer's Disease	05/01/19-04/30/23 \$60,000 Total Project Costs
David Weidman (Site PI) 5 P30 AG019610 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center – Clinical Core	7/1/2018 - 6/30/2021 \$68,253 Annual Total Costs
David Weidman (Site PI) 5 P30 AG019610 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center – Outreach and Recruitment Core	7/1/2018 - 6/30/2021 \$48,870 Annual Total Costs
David Weidman (Project PI) Arizona Alzheimer's Research Consortium (Reiman) AZ DHS via AARC Native American Outreach Program	7/1/2019-6/30/2020 \$75,000 Annual Total Costs
Thomas Beach (Co-I; Core Leader) P31AG19610 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center	7/1/16-6/30/21 \$116,603 Annual DC
Thomas Beach (PI) Grant Michael J. Fox Foundation for Parkinson's Research Systemic Synuclein Sampling Study (S4)	1/1/20-12/31/20 \$62,669 Annual DC

Thomas Beach (Co-I) Grant	4/1/19-3/31/20
Carl T. Hayden Medical Research Foundation (Migrino) Discovering novel mechanisms for aging-related dementia: probing medin and abeta vasculopathy	\$4,476 Annual DC
Thomas Beach (Site Leader) Navidea Biopharmaceuticals	4/1/14-present
Dr. Beach is Leader of the Central Neuropathology Site for this imaging-to-autopsy Phase III clinical trial of an amyloid imaging agent for diagnostic usage.	\$100,000 Annual DC
Thomas Beach (Site Leader) Avid Radiopharmaceuticals	9/1/15-present
Dr. Beach is Leader of the Central Neuropathology Site for this imaging-to-autopsy Phase III clinical trial of a tau PET imaging agent for diagnostic usage.	\$250,000 Annual DC
Thomas Beach (Project PI) Arizona Alzheimer's Research Consortium (AARC) AAC via AZ DHS	7/1/19-6/30/20 \$160,000 Annual DC
Developing a Shared Resource of CSF, Plasma, Serum, PBMC samples from Arizona's Longitudinal Brain and Body Donation and APOE4 Gene Dose Program	
Thomas Beach (Project Co-PI) Arizona Alzheimer's Research Consortium (AARC) AAC via AZ DHS	7/1/19-6/30/20 \$160,000 Annual DC
Towards Single-Cell Analysis in Human Brain Neurodegenerative Disease: A Pilot Study	
Thomas Beach (Co-I) Grant (Reiman) Nomis Foundation	7/1/17-6/30/20 \$116,603 Annual DC
A Public Resource of RNA Sequencing Data from Alzheimer's and Aging Brain	
Thomas Beach (Co-I) Arizona Alzheimer's Research Consortium (AARC) AAC via AZ DHS	7/1/19-6/30/20 \$144,000 Annual DC
A Human Brain Single-Cell Suspension Resource	
Thomas Beach (Co-I) Arizona Alzheimer's Research Consortium (AARC) AZDHS via AARC	7/1/19-6/30/20 \$36,000 Annual DC
Clinicopathological Study Initiation for Incidental REM Sleep Behavior Disorder in Sun City	
Thomas Beach (Co-I) 5R01NS110188-02 (LaVoie) NIH via Brigham & Women's/HMS	7/1/19-6/30/20 \$19,841 Annual DC
Pathologic LRRK2 signaling in Familial and Idiopathic Parkinson's Disease	
Thomas Beach (Co-I) 1R01NS112203-01 (Yacoubian) NIH via University of Alabama-Birmingham	9/15/19-5/31/20 \$17,156 Annual DC
14-3-3 phosphorylation in Parkinson's Disease	

Thomas Beach (Co-I) R01 NIH via University of CA-Irvine PET Imaging Agents for a4b2 Nicotinic Receptors	12/1/18-11/30/21 \$25,000 Annual DC
Thomas Beach (Co-I) NIH R33 NIH via Boston University The B cell repertoire as a window into the nature and impact of the lung virome	5/1/9-4/30/22 \$12,000 Annual DC
Geidy Serrano (Co-PI) AARC (Reiman, Project PIs: Serrano, Beach) AZ DHS via AARC A Human Brain Single-Cell Suspension Resource	7/1/19– 6/30/20 \$144,000 Annual DC
Geidy Serrano (PI) 2019 Grant Michael J. Fox Foundation Characterization of isolated human astrocyte population in aging and Lewy body pathology	10/1/19-3/31/21 \$118,716 Annual DC
Christine Belden (Neuropsychologist) 5 P30 AG019610 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center – Clinical Core	8/15/16 - 6/30/21 \$12,513,020
David Shprecher (Site Leader) 3R34AG056639-02S1 NIH via Washington University (Ju) Neuroprotective Treatment Trial Planning in REM Sleep Behavior Disorder	9/01/19 - 4/30/20 \$25,280
David Shprecher (Project PI) Arizona Alzheimer's Research Consortium (Reiman) AZ DHS via AARC (Shprecher, Project PI) Laying the Groundwork for Prodromal Lewy Body Dementia Research in REM	7/01/19 - 6/30/20 \$40,000
Alireza Atri (Clinical Site PI/Co-I) 5P30 AG019610 (Reiman) NIH/NIA via ASU Arizona Alzheimer's Disease Core Center	7/1/11-6/30/21 \$95,711 Annual DC
Alireza Atri (Clinical Site PI/Co-I) P30 AG19610-19S1A1 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center – Brain Imaging and Fluid Biomarker Core (BIFB Core)	9/15/18 -6/30/21 \$1,242,362
Alireza Atri (Co-I) AARC (Reiman, Project PI: Beach) AZ DHS via AARC Developing a Shared Resource of CSF, Plasma, Serum , PBMC samples from Arizona's Longitudinal Brain and Body Donation and APOE4 Gene Dose Program	7/1/19-6/30/20 \$185,000 Annual DC

Alireza Atri (PI) AARC (Reiman, Project PI: Atri) AZ DHS via AARC Enhancing Clinical and Biological Characterization of The Longevity Cohort Study: Global Staging and Biospecimen Banking	7/1/19-6/30/20 \$85,000 Annual DC
Alireza Atri (Co-PI) AARC (Reiman, Project PI: Goldfarb) AZ DHS via AARC Advancing Lumbar Puncture in ADRD Research	7/1/19-6/30/20 \$25,000 Annual DC
Alireza Atri (Site PI) U24 AG057437 (Aisen) NIH/NIA via USC (ATRI) Alzheimer's Clinical Trial Consortium	12/02/17-11/30/22 \$896,667 Total Project Costs
Alireza Atri (Site PI) R01 AG053798 (Aisen) NIH/NIA via USC (ATRI) Global Alzheimer's Platform Trial-Ready Cohort for Preclinical/Prodromal Alzheimer's Disease	05/01/19-04/30/23 \$60,000 Total Project Costs
Alireza Atri (Site PI) Grant (Atri) Global Alzheimer's Platform Foundation	01/01/20-12/31/20 \$100,000 Total Project Costs
Sylvia E. Perez (PI) 19-500-275-30-09 Arizona Alzheimer's Consortium Splicing and tau pathology in Down syndrome and Alzheimer's disease	07/01/19-06/30/20 \$72,727
Sylvia E. Perez (Collaborator) P01 AG014449 (Mufson) NIH-NIA Neurobiology of mild cognitive impairment in the elderly	09/01/97-01/31/25
Rita Sattler (Co-PI) RO1 Supplement award (Zarnescu) NIH NINDS RNA Dysregulation in neurodegeneration	11/01/19-10/31/20 \$91,000
Rita Sattler (Co-I) SBIR (Sucholeiki (Aquilus Pharma)) NIH NINDS Matrix Metalloproteinase 2/9 inhibitor for the treatment of ALS	09/15/19-08/31/20 \$25,000
Rita Sattler (PI) Award Department of Defense Genotypic and Phenotypic Examination of Disease Pathogenesis in C9orf72 FTD	07/01/19-06/30/22 \$250,000

Rita Sattler (PI) AARC	07/01/19-06/30/20
Arizona Alzheimer's Research Consortium/Barrow Neurological Institute	\$80,000
Nucleocytoplasmic trafficking deficits of ADAR2 and RNA editing aberrations in Alzheimer's disease	
Rita Sattler (PI) Award (Sattler/Bowser/Mufson/Shefner) Fein Foundation	01/01/19-12/31/20 \$150,000
Studies of disease mechanisms and biomarker development for FTD	
Rita Sattler (Co-PI) Award (Zarnescu) Department of Defense	07/01/18-06/30/20 \$75,000
Small molecules targeting TDP43-RNA interaction in ALS	
Rita Sattler (Co-PI) Award (Dracheva) Department of Veteran's Affairs	07/01/17-06/30/21 \$70,000
The role of ADAR2-associated RNA editing in pathogenesis of ALS	
Rita Sattler (Mentor) Graduate Student Scholarship (Moore) Barrow Neurological Foundation	01/12/18-12/31/19 \$10,000
Molecular mechanisms of RNA editing deficits in C9orf72	
Rita Sattler (Mentor) Postdoctoral Fellowship (Lorenzini) Barrow Neurological Foundation	07/01/19-06/30/20 \$85,000
Role of microglial cells in C9orf72-mediated hiPSC cortical neuron synaptic dysfunction	
Ashley Stokes (PI) AZ Alzheimer's Disease Consortium Award AAC via AZ DHS	07/01/19-06/30/20 \$80,000
Multi-Scale MRI Assessment of Neurovascular Factors Associated with AD	
Ashley Stokes (PI) AZ Alzheimer's Disease Consortium Award AAC via AZ DHS	07/01/19-06/30/20 \$80,000
Development of a Multi-Scale MRI Method for Preclinical Validation of Hemodynamic Factors Associated with AD	
Ashley Stokes R01 CA158079 (Quarles) NIH/NCI	09/16/11-07/31/21 \$207,500
MRI Assessment of Tumor Perfusion, Permeability and Cellularity	
Ashley Stokes R01 CA213158-01 (Quarles) NIH/NCI	07/01/17-06/30/21 \$175,000
Establishing the validity of brain tumor perfusion imaging	

Ashley Stokes P30 AG019610-20 (Reiman) NIH/NIA ARIZONA ALZHEIMER'S DISEASE CORE CENTER (ADCC): Brain Imaging and Fluid Biomarkers (BI-FB) Core (Core G) Core PI: Alexander	07/01/18-06/30/23 \$1,229,916
Ashley Stokes Award (Alhilali) ASFNR Evaluation of a Single Bolus, Multi-Echo Dynamic Susceptibility Contrast Protocol in Patients with Glioblastoma	07/01/19-06/30/20 \$60,000
Joseph Scheeren (Contact PI) Food and Drug Administration 2U18FDA005320-06 CRITICAL PATH TO PUBLIC PRIVATE PARTNERSHIPS	09/01/19-08/31/20 \$8,595,141
Layla Al-Nakkash (PI) Diabetes Action and Research Foundation High fat diet-induced diabetes is abolished by combined genistein and exercise treatment: identifying the mechanisms	01/01/19-06/30/20 \$10,000
Layla Al-Nakkash (PI), Tom Broderick & Minsub Shim (Co-PIs) Arizona Alzheimer's Consortium Diabetic obesity results in cognitive impairment: evaluation of the relationship between inflammation, and senescence in the gut-brain axis and the response to genistein and exercise treatment	07/01/10-06/30/20 \$24,896
Layla Al-Nakkash (PI) Midwestern University-Intramural Grant Assessment of the effects of genistein and exercise on intestinal physiology in high fat-high sugar fed mice	07/01/19-06/30/20 \$5,000
Layla Al-Nakkash (Co-PI) Midwestern-One Health Research Stimulus Award Effects of calcitriol on leukocyte cytokine production in dogs with type 1 diabetes mellitus	07/01/19-06/30/20 \$10,000
Layla Al-Nakkash (Consultant) NIH-R15 Physical activity as a therapeutic intervention in endometriosis	04/01/18-03/31/21 \$470,124
Delrae Eckman (PI) Arizona DHS, Arizona Biomedical Research Commission Cerebrovascular Dysfunction and Cognitive Decline in Aging APOE2, APOE3 and APOE4 Targeted-Replacement Mice	04/01/18-03/31/21 \$225,000
Mitra Esfandiarei (Co-I) NIH R15 Targeting Endothelial Dysfunction in a Genetic Mouse Model of Aortic Aneurysm: Implications for Prevention and Therapy	01/15/19-12/31/21 \$441,048

Vanthida Huang (PI) Midwestern University CPG The goal of this study is to evaluate the phenotypic and genotypic relatedness of <i>Staphylococcus aureus</i> and <i>Pseudomonas aeruginosa</i> isolated from veterinary and human origin using pulse-field gel electrophoresis	07/01/19-06/30/20 \$10,720
Nafisa Jadavji (PI) American Heart Association Research Enhancement Award Identification of developmental factors involved in ischemic stroke outcomes in adulthood and old age	01/01/20-12/31/21 \$152,735
Garilyn Jentarra (PI) and T. Bucky Jones (Co-PI) Arizona Alzheimer's Consortium Following the Evidence: The Role of Microbes in the Development of Alzheimer's Disease	07/01/19-06/30/20 \$40,183
Garilyn Jentarra (PI) Midwestern University-Intramural Grant Is the tau hyperphosphorylation observed in Alzheimer's disease an anti-microbial response?	07/01/19-06/30/20 \$5,000
Ashlesh Murthy (PI) NIH/NIAID 2R15AI101920-03 Mechanism(s) of CD8 T cell-mediated Chlamydia-induced reproductive pathology	07/01/19-06/30/22 \$450,000
Ashlesh Murthy (PI) USDA-NIFA USDA Animal Health and Research Disease Grant	10/01/18-09/30/20 \$25,604
Ashlesh Murthy (PI) USDA-NIFA USDA Animal Health and Research Disease Grant	10/01/19-09/30/20 \$25,604
Minsub Shim (PI) NIH/NCI R15CA246429 Cyclooxygenase-2 signaling in cell senescence and its role in chemotherapy-induced long-term adverse sequelae.	12/01/19-11/30/22 \$450,000
Minsub Shim (PI), Layla Al-Nakkash & Tom Broderick (Co-PIs) Arizona Alzheimer's Consortium Exercise-mediated mitigation of cellular senescence as a peripheral control mechanism for Alzheimer's disease risk.	07/01/19-06/30/20 \$28,702
Mark Swanson (PI) and Nancy Bae (Co-PI) Arizona Alzheimer's Consortium Studies on the effects of the telomere protection protein RAP1 and the epsilon isoform of Glial Fibrillary Acidic Protein on amyloid peptides produced by gamma-secretase.	07/01/19-06/30/20 \$6,219
Charles Veltri (PI) Center for Plant Science and Health Pharmacological and Phytochemical Investigations of Kratom ( <i>Mitragyna speciosa</i> Korth.) in the Nematode <i>Caenorhabditis elegans</i> .	12/01/19-11/30/20 \$9,978

J Gregory Caporaso (Investigator)  
478478 / ADHS18-198857 (Duca) 04/01/18 -03/31/21  
University of Arizona / Arizona Biomedical Research Commission \$17,604  
Role of the Small Intestine in the Prebiotic Treatment/or Obesity

J Gregory Caporaso (PI)  
478478 / ADHS18-198857 (Caporaso) 04/01/18-03/31/21  
University of Arizona / Arizona Biomedical Research Commission \$38,225  
Role of the Small Intestine in the Prebiotic Treatment/or Obesity

J Gregory Caporaso (PI)  
2019-207342 (Caporaso) 12/01/19-11/30/20  
Chan Zuckerberg Initiative / Silicon Valley Community Foundation \$132,391  
Advancing microbiome research through QIIME 2 user and developer community development

J Gregory Caporaso (Investigator)  
G-2019-12432 (Stachurski) 12/15/19-10/31/22  
Alfred P. Sloan Foundation / The Australian National University \$59,535  
Document Creation and Publishing Tools for Next-Generation Scientific Textbooks

Emily Cope (PI), J Gregory Caporaso (PI)  
1R15AI147148-01A1 (Cope, Caporaso) 07/01/19-06/30/22  
NIH/NIAID \$72,420  
Determining the Role of the Upper and Lower Airway Microbiota as Drivers of Concomitant  
Inflammatory Responses in patients with Chronic Rhinosinusitis and Asthma.

J Gregory Caporaso (Project PI)  
5U54CA143925 (Ingram) 09/01/19-08/31/24  
NIH/NCI \$60,000  
The Partnership for Native American Cancer Prevention.

J Gregory Caporaso (Investigator)  
1U54MD012388 (Baldwin, Stearns) 07/01/17-06/30/22  
NIH / NIMHD \$435,353  
Southwest Health Equity Research Collaborative (SHERC)

J Gregory Caporaso (Investigator)  
2017-67013-26255 / F0003750302014 (Kaplan) 04/01/20-03/31/22  
National Institute of Food and Agriculture / Purdue \$40,012  
Optimizing Plant-Soil Feedbacks for High Intensity Crop Production Systems

Emily Cope (PI), J Gregory Caporaso (PI)  
ADHS 14-052688 / Grant # CTR040636 07/01/19-06/30/20  
Arizona Alzheimer's Research Center, Inc \$50,000  
Longitudinal analysis of the gut microbiome-brain axis in Alzheimer's Disease

J Gregory Caporaso (PI)  
1565100 05/01/16-04/30/20  
National Science Foundation \$525,795  
Extensible, reproducible and documentation-driven microbiome data science.

J Gregory Caporaso (Co-I) APP1085372 National Health and Medical Research Council (Huttley) Robust bioinformatics for predicting bacterial pathogens from microbiome sequencing	01/01/15-01/30/20 \$137,868
Emily Cope (PI) TRIF-SPA 2.0 (Cope) NSF-CLP Postdoctoral scholar award: Upper and lower airway microbiota in asthma	07/01/17-06/30/19 \$160,000
Emily Cope (Other) Flinn Foundation Research Grant (Keim/Lal/LaBaer MPI) Flinn Foundation Characterization of the Chronic Rhinosinusitis (CRS) Microbiome-Host Interaction for Microbiota-Directed Probiotic Therapy	01/01/16-12/30/19 \$1,200,000
Emily Cope (Co-I) 1U54MD012388-01 (Baldwin/Stearns-MPI) SHERC Pilot grants program (Pilot: Koppisch, Co-I: Cope) NIH/NIMHD Novel Ionic Liquid Formulations to Combat Diabetic Foot Ulcer Infections	02/01/18-01/31/20 \$59,892
Emily Cope (PI) 1U54MD012388-01 (Baldwin/Stearns-MPI) SHERC Pilot grants program (PI: Cope) NIH/NIMHD Addressing asthma health disparities through diet-based modification of the gut-microbiome airway axis	02/01/18 - 01/31/20 \$59,997
Emily Cope (PI) Flinn Foundation Research Grant (Cope/Rank MPI) Project #2188 Precision Treatment of Asthma Through Targeted Manipulation of the Gut Microbiome Lung Axis	01/01/19-12/30/21 \$100,000
Emily Cope (PI) Elizabeth Nash Postdoctoral Fellowship Cystic Fibrosis Research Inc. A Multi-'Omic Approach to Evaluate Concurrent Sinus and Pulmonary Disease in Cystic Fibrosis	07/01/19-06/30/21 \$120,000
John Fryer (PI) R01NS094137 (Fryer) NIH/NINDS The role of Clusterin in cerebral amyloid angiopathy	09/01/15-08/31/20
John Fryer (PI) R21AG057997 (Kang/Fryer) NIH/NIA Impact of APOE3 and APOE4 on the peripheral immune system in Alzheimer's disease	02/01/18-01/31/20

John Fryer (PI) R01AG06211 (Fryer/Liu) NIH/NIA Microglial apoE in neuroinflammation and Alzheimer's disease	2018 - 2023
John Fryer (PI) R01AG062077 (Fryer/Petrucci) NIH/NIA Novel genetic modifiers of C9orf72 and Tau toxicity	2019 - 2024
John Fryer (PI) R56AG062556 (Springer/Fryer) NIH/NIA Selective autophagy in Alzheimer's disease and related dementias	2019 - 2020
John Fryer (Project PI) U54NS110435 (Ross/Fryer/Chang) NIH/NINDS Lewy Body Dementia CWOW, Project 1: Omics driven network analysis in LBD	2019 – 2024
John Fryer (PI) Coins for Alzheimer's Trust The role of microglial lipid signaling in Alzheimer's disease pathogenesis	2019 – 2021
John Fryer (PI) Cure Alzheimer's Foundation curealz.org The role of Clusterin in tau pathology	02/01/20-01/31/22
John Fryer (PI) Mayo Clinic Alzheimer's Disease Center pilot grant Single-cell/nucleus transcriptional signatures underlying Alzheimer's disease pathology	07/01/19-06/30/22
Richard Caselli (PI/Core Leader) P30 AG019610 (Reiman) NIH/National Institute on Aging Arizona Alzheimer's Disease Core Center Core B: ADCC Clinical Core	07/01/16-06/30/21 \$191,728
Richard Caselli (Consortium PI) P30 AG019610 (Reiman) NIH/National Institute on Aging Core C: ADCC Data Core: Alzheimer's Disease Core Center	07/01/16-06/30/21 \$18,962
Richard Caselli (Consortium PI) P30 AG019610 (Reiman) NIH/National Institute on Aging Core A: ADCC Admin Core: Alzheimer's Disease Core Center	07/01/16-06/30/21 \$18,962
Richard Caselli (PI) R01AG031581 (Reiman/Caselli) NIH/NIA PET, APOE, & the Preclinical Course of Alzheimer Disease	05/01/08-03/31/21 \$126,358

Richard Caselli (Co-I) R01AG054048 (Sierks) NIH/NIA Protein Variants as Blood Based Biomarkers for Diagnosing and Staging Alzheimer's Disease	07/01/16-06/30/21 \$19,392
Richard Caselli (PI) ADHS12-010553 (Caselli) Arizona Department of Health Services Normal and Pathological Aging (Preclinical Alzheimer's Disease)	07/01/19-06/30/20
Dona Locke (PI) Ralph C. Wilson Foundation Development Fund HABIT Registry	09/01/17-Present
Dona Locke (Co-I) ADHS12-010553 (Caselli) Arizona Department of Health Services Normal and Pathological Aging (Preclinical Alzheimer's Disease)	07/01/19-06/30/20
Matthew Huentelman (Co-I) RO1 AG049465 (Barnes) NIH/NIA Neural System Dynamics and Gene Expression Supporting Successful Cognitive Aging	08/01/14-03/31/20 \$162,203
Matthew Huentelman (PI) AAC - DHS (Huentelman) (Reiman) State of Arizona, DHS AARC FY 20: Alzheimer's Projects	07/01/19-06/30/20 \$100,000
Matthew Huentelman (Co-I) 1 RO1 AG049464 (Coleman/Barnes/Alexander) NIH/NIA Epigenetic, Neuroimaging and Behavioral Effects of Hypertension in the Aging Brain	08/01/14-07/31/20 \$178,013
Matthew Huentelman (Co-PI) Grant#20170715 (Padilla) Aging Mind Foundation Early Onset Alzheimer's Disease Genomic Study	09/01/17-11/20/20 \$32,441
Matthew Huentelman (Co-I) P30 AG019610 (Reiman) NIH/NIA Arizona Alzheimer's Disease Core Center	07/01/16 - 06/30/21 \$12,190
Matthew Huentelman (Co-I) UG30D023313 (Deoni) NIH The Developing Brain: Influences and Outcomes	09/21/16-08/31/21 \$70,000
Matthew Huentelman (Co-I) R01AG054180 (Kaczorowski)	05/15/17-04/30/22

NIH Systems Genetics of Cognitive Aging and Alzheimer's Disease	\$10,028
Matthew Huentelman (Co-I) R56HL141165 (Hale)	09/20/18-08/31/23
NIH Identifying a Pathogenic Fibroblast Subpopulation to Target for Protection Against Cardiac Fibrosis	\$48,416
Matthew Huentelman (Co-I) W81XWH1910534 (Schwedt)	09/01/19 -08/31/23
DOD A multidisciplinary translational approach to investigate the mechanisms, predictors and prevention of persistent post traumatic headache	\$181,830
Winnie Liang (Co-I) Contract (Keats)	07/01/11 -06/30/20
MMRF Longitudinal, Observation Study in Newly Diagnosed Multiple Myeloma (MM) Patients to Assess the Relationship between Patient Outcomes, Treatment Regimens and Molecular Profiles (The MMRF Longitudinal Study)	\$1,364,526
Winnie Liang (Co-I) W81XWH-16-TSCRIP-IDA (Narayanan)	12/01/16-11/30/19
DoD TS160074:Phenotypic Variability in Tuberous Sclerosis Complex (TSC)	\$150,000
Winnie Liang (Co-I) Grant (Reiman)	07/01/17-06/30/21
NOMIS Foundation	\$1,133,608
Winnie Liang (Co-PI) Grant (Hendricks/Cowey)	09/01/17-08/31/19
Baylor Scott & White Research Institute (BSWRI) Dissecting the role of hTERT across melanoma subtypes: Exploiting a cancer hallmark to develop a novel targeted treatment	\$223,000
Winnie Liang (Co-I) 1U01CA224153 (London)	09/30/17-08/31/22
NIH/Tufts University Subaward Precision Medicine for Pet Dogs with Lung Cancer	\$46,541
Winnie Liang (PI) Grant (Huentelman)	07/01/19 – 06/30/20
Arizona Alzheimer's Research Consortium AARC FY 19: Alzheimer's Projects	\$100,000
Winnie Liang (Co-I) R01 CA223481 (Murtaza)	08/15/18-07/31/23
NIH/NCI	\$179,316

Individualized monitoring of treatment response and resistance in patients with metastatic melanoma

Winnie Liang (Co-I) R01CA195670 (Weissman, Huntsman, Trent) NIH/NCI The Tumor Suppressor Role of SMARCA4 in SCCOHT	06/01/15-05/31/20
Kendall Van Keuren-Jensen (Co-I) Grant (Craig) Michael J Fox Foundation for Parkinson's Research Identification of RNA Isoform and Splicing Based Biomarkers For Parkinson's Disease	05/01/19-04/30/22 \$60,000
Kendall Van Keuren-Jensen (Multi-PI) UG3UH3 (Das) NIH Molecular dissection and imaging of extracellular vesicles to define their origin and targets	07/01/19-06/30/23 \$463,700
Kendall Van Keuren-Jensen (PI) Grant Foundation for the National Institutes of Health Data QC and analysis for BioFIND and PDBP	03/01/19-02/28/20 \$289,940
Kendall Van Keuren-Jensen (Consortium PI) UG3UH3 (Laurent) NIH Development and application of a scalable workflow for immunomagnetic separation of exRNA carrier subclasses and molecular analysis of their cargo.	07/01/19-06/30/23 \$260,000
Kendall Van Keuren-Jensen (Multi-PI) UG3UH3 (Raffai) NIH P.R.I.S.M. : Purification of exRNA by Immuno-capture and Sorting using Microfluidic	07/01/19-06/30/20 \$369,468
Kendall Van Keuren-Jensen (PI) W81XWH1910277 Dept of the Army (USA MRC) Genotypic and phenotypic examination of disease pathogeneis in C9orf72 FTD	07/01/19-06/30/22 \$587,368
Kendall Van Keuren-Jensen (Multi-PI) Grant Michael J Fox Foundation for Parkinson's Research Extracellular vesicles from urine are enriched in brain transcripts and have potential as noninvasive biomarkers	04/16/19-04/15/21 \$170,000
Kendall Van Keuren-Jensen (PI) Grant 977871 Sidell-Kagan Foundation Advancing prevention of Alzheimer's disease: Extracellular RNAs as candidates for monitoring Alzheimer's patients	06/01/17-05/31/20 \$150,000

Kendall Van Keuren-Jensen (PI) Grant ID: 17491 Michael J Fox Foundation for Parkinson's Research Collaboration and Advisory role for the Industry LRRK2 Detection Consortium: CSFExosome Study	02/14/19-02/13/20 \$19,029
Kendall Van Keuren-Jensen (Co-I) Grant 15065 (Singleton-NIH) Michael J Fox Foundation for Parkinson's Research The Foundational Data Initiative	05/24/18-05/23/20 \$158,726
Kendall Van Keuren-Jensen (Co-I) Grant 14539 (Cookson-NIH) Michael J Fox Foundation for Parkinson's Research LRRK2 Biology Consortium Program	06/08/18-06/04/20 \$80,000
Kendall Van Keuren-Jensen (PI) Grant ID: 12749.01 Michael J Fox Foundation for Parkinson's Research RNAseq and miRNAseq in PPMI whole blood samples (2nd)	04/01/17-08/31/19 \$90,568
Ahern, Geoff (co-I; PI: Reiman) NIH/NIA P30 AG019610 Arizona Alzheimer's Disease Core Center (UA Clinical Core)	07/01/16-06/30/21 \$43,084 Annual DC
Ahern, Geoff (PI) Novartis Pharmaceutical Corporation A Randomized, Double-blind, Placebo-controlled, Two cohort Parallel Group Study to Evaluate the Efficacy Of Cad106 and Cnp520 in Participants at Risk for the Onset Of Clinical Symptoms of Alzheimer's Disease.	2019 – present \$172,970
Alexander, Gene (multi-PI: Alexander, Bowers, Woods) NIH/NIA R01AG046587 Revitalizing Cognition in Older Adults at Risk for Alzheimer's Disease with Near-Infrared Photobiomodulation	08/01/19 – 04/30/24 \$378,525 Annual TC
Alexander, Gene (multi-PI: Reiman, Caselli) NIH/NIA RO1 AG031581 Brain Imaging, APOE & the Preclinical Course of Alzheimer's disease	05/01/18 – 03/31/20 \$14,630 UA Annual TC
Alexander, Gene (PI's: Coleman, Barnes, Alexander; co-I's: Billheimer, Huentelman, Trouard) NIH/NIA 1 RO1 AG049464 Epigenetic, Neuroimaging and Behavioral Effects of Hypertension in the Aging Brain	08/01/14 – 05/31/20 \$458,236 Annual DC
Alexander, Gene (PI, UA Sub; co-I's: Trouard, Hishaw, Allen) NIH/NIA RO1 AG054077 Augmenting Cognitive Training in Older Adults	09/01/16 – 06/30/21 \$255,362 Annual TC
Alexander, Gene (co-I; PI: Su) NIH/NIA RO1 AG055020	09/01/16 – 06/30/21 \$85,653 Annual TC

Ultra-sensitive and Label-free Detection of Alzheimer's Disease Biomarkers

Alexander, Gene (co-I, PI: Reiman) NIH/NIA P30 AG019610 Brain Imaging and Fluid Biomarkers Core	08/01/2017 – 04/30/20 \$304,037 UA Total DC
Alexander, Gene (Co-I, PI: Reiman) NIH/NIA P30 AG019610 Arizona Alzheimer's Disease Core Center	7/1/16 - 6/30/21 \$18,950 Annual TC
Alexander, Gene (Multi-PI: Bowers, Alexander, Woods) McKnight Brain Research Foundation A Pilot Intervention with Near Infrared Stimulation: Revitalizing Cognition in Older Adults	10/01/19 – 9/30/21 \$60,000 Total DC
Alexander, Gene (UA PI; PI Williamson) McKnight Brain Research Foundation Transcutaneous Vagal Nerve Stimulation and Cognitive Training to Enhance Cognitive Performance in Healthy Older Adults	05/01/18 – 04/30/20 \$60,000 Total DC
Alexander, Gene (PI; co-I's: Raichlen, Ahern, Beach, Caselli, Su, Huentelman, Klimentidis, Reiman, Ryan, Trouard) State of Arizona, DHS Grant Influence of Physical Activity on Brain Aging and the Risk for Alzheimer's Disease	07/01/19 – 06/30/20 \$57,463 Annual DC
Andrews-Hanna, Jessica (UA PI; Bryan PI) NIH/NIA RO1 AG043452 Enhancing Function in Later Life: Exercise and Function Network Connectivity	06/15/14 – 06/30/19 \$15,232 Annual TC
Andrews-Hanna, Jessica (PI; co-I's: Grilli, O'Connor) NIH/NIA P30 AG019610 Uncovering Neurocognitive Links between Alzheimer's Disease and Depression in Mid-Life to Early Aging	07/01/18 – 06/30/19 \$30,000 Total DC
Andrews-Hanna, Jessica (PI; co-I's: Magrino; Maddine) State of Arizona, DHS Grant Real-world Markers and Neural Mechanisms of Alzheimer's disease risk in cognitively normal older adults	07/01/19 – 06/30/20 \$22,143 Annual DC
Arce, Fernando Teran (PI; co-I's: Grilli, Huentelman, Mehl, Ryan) State of Arizona, DHS Grant Role of medin membrane pores in AD and vascular-dementia-related vasculopathy	07/01/19 – 06/30/20 \$26,604 Annual DC
Barnes, Carol (PI) NIH/NIA 1 R01 AG050548 Cell Assemblies, Brain Adaptation and Cognitive Brain	09/1/15 – 05/31/20 \$516,626 Annual TC
Barnes, Carol (PI; co-I: Ekstrom) NIH/NIA 1 RO1 AG003376 Neurobehavioral Relations in Senescent Hippocampus	01/01/16 – 11/30/20 \$734,165 Annual TC

Barnes, Carol (PI's: Barnes, Huentelman; co-I: Okuno) NIH/NIA 1 RO1 AG048907 CATT: Development and Application of a Neuronal Cell Activity-Tagging Toolbox	09/30/14 – 05/31/20 \$251,270 Annual DC
Barnes, Carol (PI) (co-I's: Alexander, Billheimer, Huentelman, Trouard) NIH/NIA RO1 AG049465 Neural System Dynamics and Gene Expression Supporting Successful Cognitive Aging	08/01/14 – 3/31/20 \$734,176 Annual TC
Barnes, Carol (PI) co-I: Reiman, Coleman, Bimonte-Nelson, Huentelman) NIA/NIA T32 AG044402 Postdoctoral Training, Neurobiology of Aging and Alzheimer's Disease	05/15/16 – 04/30/21 \$260,293 Annual TC
Barnes, Carol (Co-I; PI: Reiman) NIH/NIA 5 P30 AG019610 Arizona Alzheimer's Disease Core Center Ad Hoc Review Program	08/15/17 – 06/30/20 \$15,945 Annual DC
Barnes, Carol (PI; Co-I's: Cai, Hruba, Trouard) State of Arizona, DHS Grant Age-related Specific Changes in Expression of Several central Melanocortin Receptor Subtypes and their Localization in Rat Brain	07/01/19 – 6/30/20 \$18,079 Annual DC
Barnes, Carol (PI; Co-I's: Hay, Guzowski) State of Arizona, DHS Grant Neuroinflammation, Aging, and Cognition: An Intervention Study	07/01/19 – 6/30/20 \$24,922 Annual DC
Brinton, Roberta (PI; co-I's: Hernandez, Rodgers) NIH/NIA RO1 AG063826 Allopregnanolone as a Regenerative Therapeutic for Alzheimer's: Phase 2 Clinical Trial	08/15/19 – 04/30/24 \$37,329,334 Total Costs
Brinton, Roberta (PI; co-I: Yin) NIH/NIA1 PO1 AG026572 Perimenopause in Brain Aging and Alzheimer's Disease	09/01/16 – 08/31/21 \$2,070,810 Annual TC
Brinton, Roberta (PI) NIH/NIA R37 AG053589 Aging and Estrogenic Control of the Bioenergetic System in the Brain	03/15/17 – 02/28/22 \$1,629,669 Total Costs
Brinton, Roberta (PI; co-I: Rodgers) NIH/NIA UO1 AG047222 Allopregnanolone as a Regenerative Therapy for Alzheimer's: FDA-Required Toxicology	06/15/14 – 02/28/19 \$215,348 Annual TC
Brinton, Roberta (PI; co-I: Yin) NIH/NIA R01 AG059093 Metabolic Networks and Pathways Predictive of Sex Differences in AD Risk and Responsiveness to Treatment	08/01/18 – 06/30/23 \$694,000 Total Costs
Brinton, Roberta (PI; co-I: Yin; Rodgers) NIH/NIA R01 AG057931	09/01/18 – 08/31/23 \$3,737,620 Total Costs

Sex Differences in the Molecular Determinants of Alzheimer's Disease Risk: Prodromal Endophenotype

Brinton, Roberta (PI) 09/01/18 – 08/31/23  
NIH/NIA T32 AG061897 \$1,334,834 Total Costs  
Translational Research in AD and related Dementias (TRADD)

Brinton, Roberta (PI) 12/04/18 – 11/01/20  
Women's Alzheimer's Movement \$100,000 Total Costs  
Does Hormone Replacement Therapy, alone or in combination with other therapies, reduce a woman's risk for Alzheimer's Disease in women who have been treated for breast cancer?

Brinton, Roberta (PI; co-I: Rodgers) 11/01/17 – 12/31/20  
Alzheimer's Drug Discovery Foundation \$150,000 Total DC  
Allopregnanone Novel Patentable Formulations to Advance Commercialization

Brinton, Roberta (PI) 04/10/19 – 03/31/21  
Alzheimer's Association \$248,280 Annual DC  
Advancing Allopregnanone as a Regenerative Therapeutic for Alzheimer's

Chen, Nan-kuei (PI; co-I's: Chou, Saranathan, Guzman, Stokes) 07/01/19 – 06/30/20  
State of Arizona, DHS Grant \$24,960 Annual DC  
Characterization of dynamic CBF and BOLD signals of the hippocampus with amnesic mild cognitive impairment

Chen, Nan-kuei (PI, co-I's: Guzman, Rapcsak, Trouard) 07/01/18 – 03/31/23  
NIH/NINDS R01 NS102220 \$285,904 Annual DC  
Development of High-Speed and Quantitative Neuro MRI Technologies for Challenging Patient Populations

Chen, Nan-kuei (UA Subcontract PI) 03/05/18 – 01/31/23  
NIA R01 DA045565 (Meade, Duke University, PI) \$15,218 Annual DC  
MRI data fusion to investigate effects of drug abuse on HIV neurological complications

Chou, Ying-hui (co-I; PI: Killgore) 09/01/20 – 08/31/22  
United States Army Medical Research Acquisition Activity \$199,997 Annual DC  
Transcranial Magnetic Stimulation of the Default Mode Network to Improve Sleep

Chou, Ying-hui (PI;) 07/01/19 – 06/30/20  
co-Is: Ryan, Rapcsak, Chen, Kuo, Saranathan \$35,115 Annual DC  
State of Arizona, DHS Grant  
Transcranial Magnetic Stimulation for Mild Cognitive Impairment

Ekstrom, Arne (PI) 07/01/12 - 6/30/22  
NIA/NINDS NS07856 \$347,985 Annual DC  
Representation of Spatiotemporal Information in Human Episodic Memory and Navigation

Ekstrom, Arne (PI) 07/01/12 - 6/30/22  
National Science Foundation \$504,390 Total Costs  
The Neural Basis of Human Spatial Navigation in Large-scale Virtual Spaces with Vestibular Input

Fernandez, Fabian (PI) National Science Foundation \$297,000 Total Costs Programming the Aged Circadian Clock with Flashes of Precision Light	11/2019 – 11/2022
Gaffney, Kevin (PI; co-I: Rodgers) State of Arizona, DHS Grant Development of Small Molecule Dual OX1R/OX2R Agonist to Explore the Role of Orexin System in Alzheimer's Pathology	07/01/19 – 06/30/20 \$27,600 Annual DC
Grilli, Matt (PI; co-I's Andrews-Hanna, Ryan) NIH/NIA R03 AG06027 The Episodic Autobiographical Memory Hypothesis of Preclinical Alzheimer's Disease: Developing a New Approach for Early Cognitive Detection and Measurement of Alzheimer's Disease	11/01/19 – 11/01/22 \$76,750 Annual TC
Grilli, Matt (PI; co-I's: Andrews-Hanna, Mehl, Huentelman) State of Arizona, DHS Grant Improving Clinical Neuropsychological Assessment of Subtle Cognitive Decline and Mild Cognitive Impairment	07/01/19 – 06/30/20 \$22,412 Annual DC
Khanna, May (UA PI) Multi-PI: Mangravite, Brennan, Price, Schadt) Sage Bionetworks Expanding AMP-AD Target Enabling Packages to Identify Small Molecule Inhibitors that Block Protein-Protein Interactions in Cells	09/01/18 – 08/31/20 \$180,657 Total Cost
Khanna, May (UA PI) Arizona State University (NIA Subcontract) Identifying the Role of RIPK1 in Alzheimer's disease	05/01/19 – 01/14/20 \$140,810 Annual TC
Khanna, May (PI) Arizona Alzheimer's Consortium Pilot Grant Program Developing Inhibitors of RIPK1/RIPK3 Interactions for AD Therapeutics	07/01/18 – 06/30/19 \$46,050 Total Costs
Khanna, May (PI; co-I's: Huentelman) State of Arizona, DHS Grant Defining the effect of a rare mutation S305Y on CD44 causing Alzheimer's disease	07/01/19 – 06/30/20 \$27,600 Annual DC
Khanna, May (PI) Arizona Biomedical Research Commission Small Molecule Restoration of Translation Dysregulation in ALS	03/01/17 – 02/29/21 \$225,000 Total Costs
McGovern, Kathryn (PI) NIH/NIA F32 AG058440 Neuroinflammation as a therapeutic avenue for Alzheimer's disease treatment	03/01/18 – 2/21/20 \$61,610 Annual DC
Rapcsak, Steve (co-I; PI: Reiman) NIH/NIA 5 P30 AG019610 Arizona Alzheimer's Disease Core Center (UA Clinical Core)	7/1/16 - 6/30/21 \$43,073 Annual DC

Rapcsak, Stephen (PI) Masaryk University Novel Network-Based Approaches for Studying Cognitive Dysfunction in Behavioral Neurology	11/01/16 – 10/31/20 \$40,022 Total DC
Rodgers, Kathleen (PI; co-I: Brinton) NIH/NINDS R25 NS107185 Undergraduate Readyng for Burgeoning Research for American Indian Neuroscientists	07/01/19 – 06/30/24 \$203,437 Annual TC
Rodgers, Kathleen (PI; co-I's: Yin. Trouard, Stoks) State of Arizona, DHS Grant Mechanisms of Neuroprotection by Mas Agonists	07/01/19 – 06/30/20 \$27,538 Annual DC
Rodgers, Kathleen (PI; co-I: Gaffney) Department of Defense W81XWH-17-1-0599 Small Molecular Mas Agonists for the Amelioration of DMD-Associated Cardiomyopathy	09/01/17 – 08/31/20 \$863,011 Total Costs
Rodgers, Kathleen (PI; co-I: Gaffney) Department of Defense AL180143 Therapeutic Idea for the Treatment of ALS with RASRx1902	07/15/19 – 07/14/21 \$682,419 Total Costs
Rodgers, Kathleen (PI) University of California, Irvine Endothelial Progenitor Cells and Cerebrovascular Injury in the Aging Brain.	07/01/19 – 06/30/24 \$69,932
Ryan, Lee (PI) co-I's: Grilli, Andrews-Hanna, Mehl, Huentelman) State of Arizona, DHS Grant Contextual retrieval impairment in self-defining autobiographical memories as an early indicator of risk for AD	07/01/19 – 06/30/20 \$60,150 Annual DC
Ryan, Lee (PI) co-I's: Chen, Labiner, Alexander, Barnes, Trouard, Brinton, Huentelman, Beach) State of Arizona, DHS Grant Establishing pipelines data sharing and image analysis for cognitively healthy older adults at the University of Arizona	07/01/19 – 06/30/20 \$60,282 Annual DC
Saranathan, Manoj (PI) co-I's: Guzman, Chen, Chou, Ahern,) Hishaw; Reiman, Alexander, Beach State of Arizona, DHS Grant MRI and automated segmentation of thalamic nuclei for Alzheimer's Disease	07/01/19 – 06/30/20 \$17,733 Annual DC
Su, Judith (PI) NIH/NIMH R21 MH111109 Label-Free, Highly-Specific, Small Molecule Detection Using Microtoroid Optical Resonators	08/01/16 – 05/31/19 \$166,946 Annual TC
Su, Judith (PI; co-I: Alexander) NIH/NIA R03 AG055020 Ultra-Sensitive and Label-Free Detection of Alzheimer's Disease Biomarkers	07/15/17 – 04/30/20 \$76,756 Annual TC

Su, Judith (PI; co-I's: Alexander, Beach) State of Arizona, DHS Grant Determining levels of Alzheimer's biomarkers in postmortem cerebral spinal fluid and serum samples	07/01/19 – 06/30/20 \$27,188 Annual DC
Su, Judith (PI) National Science Foundation High Precision Molecular Spectroscopy and Detection Using Microtoroid Optical Resonators	08/15/18 – 07/31/20 \$100,000 Annual DC
Su, Judith (PI) Defense Threat Reduction Agency Sensitive, Selective, and Affordable Chemical Threat Sensing Using Frequency Locked Microtoroid Optical Resonators	09/01/18 – 08/31/21 \$371,899 Annual DC
Su, Judith (PI) Gordon and Betty Moore Foundation Understanding Biological Systems Using Resonator-Mediated Single-Molecule Raman Detection and Spectroscopy	03/01/19 – 02/29/20 \$56,250 Annual TC
Trouard, Ted (PI) NIH Bruker Biospec 7T Small Animal MRI Upgrade	03/01/18 – 02/29/20 \$600,000 Total Costs
Trouard, Ted (UA PI) ADHS-16-00005489 Arizona Biomedical Research Corporation Treatment of Parkinson's Disease with Enhanced Delivery of Antibody Therapy Selectively Targeting Toxic Proteins Variants	03/01/17 – 02/28/21 \$125,000 Annual TC
Trouard, Ted (co-I) NIH/NICHD R01 HD079498 Intense Physiotherapies to Improve Function in Young Children with Cerebral Palsy	05/01/14 – 10/31/20 \$227,043 Annual TC
Weinkauff, Craig (PI; co-I's: Altbach, Stokes, Ryan, Trouard) State of Arizona, DHS Grant Evaluating the Impact of Carotid Artery Endarterectomy on Cognition and Brain Function using Advanced Neuro-imaging Techniques	07/01/19 – 06/30/20 \$27,613 Annual DC
Wilson, Robert C (PI; co-I's: Alexander, Andrews-Hanna, Chou) NIA R56 AG061888 Evaluating the Neurocomputational Mechanisms of Explore-Exploit Decision Making in Older Adults	9/30/18-8/31/19 \$215,613 Annual TC
Wilson, Robert (PI) co-I's: Alexander, Andrews-Hanna, Chou, Ekstrom NIA R01 AG061888 Evaluating the Neurocomputational Mechanisms of Explore-Exploit Decision Making in Older Adults	09/01/19-08/31/24 \$1,765,250 Total Costs
Wilson, Robert (Multi-PI: Grilli, Levin, Ebner, Oliveira, Getz) McKnight Brain Research Foundation Vulnerability of Older Adults to Financial Deception Schemes	08/2018 – 09/2020 \$110,000 Total Costs

Yin, Fei (PI; co-I's: Gu, Brinton) State of Arizona, DHS Grant Effect of APOE Isoforms on Neuronal- and Astrocytic Energy Metabolism	07/01/19 – 06/30/20 \$27,042 Annual DC
Zarnescu, Daniela (PI; co-I: Khanna) NIH/NINDS R01 NS091299 RNA Dysregulation in Neurodegeneration	09/01/15 – 5/31/20 \$321,921 Annual TC
Zarnescu, Daniela (PI; co-I's: Khanna, Gokhale) United States Army Medical Research Acquisition Activity Small Molecules Targeting TDP-43 - RNA Interactions in ALS	07/01/18 – 06/30/23 \$702,930 Total Costs
Zhou, Wei (PI; co-I's: Trouard, Chou, Soman, Zaharchuk, Hsu) State of Arizona, DHS Grant Cognitive Effects of Carotid Intervention-related Brain Microinjury	07/01/19 – 06/30/20 \$23,549 Annual DC
Zhou, Wei (PI) Society for Vascular Surgery Foundation SVS Foundation Bridge Grant	10/15/18 – 08/31/20 \$50,000 Total Costs
Zhou, Wei (Site PI) CryoLife, Inc Post-market, Prospective Evaluation of Photo-oxidized Decellularized Bovine Pericardium Used as a Patch in Vascular Repair And Reconstruction Surgery	04/11/19 – 03/20/23 \$65,906 Total Costs
Zhou, Wei (PI) Cook Medical, Inc Vascular Surgery Integrated Residency Program	05/16/19 – 06/30/20 \$25,000 Total Costs
Zhou, Wei (UA PI) NIA/NINDC R01 NS097876 (Mayo Clinic Sub) Carotid Revascularization and Medical Management for Asymptomatic Carotid Stenosis Trial (CREST-2)	03/01/17 – 02/29/20 \$338,600 Total Costs
Jonathan Lifshitz (PI) Diane & Bruce Halle Foundation Translational Neurotrauma Research Program	11/15/13-11/14/20 \$500,000
Jonathan Lifshitz (Co-PI) W81XWH-17-1-0473 (Migrino) Department of Defense USAMRAA / Carl T Hayden Medical Research Foundation Probing the Mechanistic Role of Vascular Dysfunction & Vascular Inflammation in TBI Cognitive Dysfunction	08/01/17-07/31/20 \$295,110
Jonathan Lifshitz (Co-PI) R01NS100793 (Thomas) National Institutes of Health Electrochemical Assessment of Behaviorally Relevant Circuit Function After TBI	12/15/17-11/30/22 \$310,034

Jonathan Lifshitz (Mentor)  
NIH R25 NS107188 (Neisewander) 07/01/18-06/30/23  
NINDS National Institutes of Health: \$1,270,790  
Workforce Inclusion in Neuroscience through Undergraduate Research Experience - WINURE

Jonathan Lifshitz (PI)  
VA Merit I01 RX002472 03/01/19-02/28/23  
U.S. Dept. of Veterans Affairs \$278,533  
Brain Injury Rehabilitation Modality, Regulation, & Structural Plasticity

Jonathan Lifshitz (Co-PI)  
Grant 01/01/19-12/31/20  
Kemper & Ethel Marley Foundation \$796,838  
Maricopa County Collaborative on Concussions in Domestic Violence (MC3DV)

Jonathan Lifshitz (PI)  
Arizona Alzheimer's Consortium (AAC) 07/01/19-06/30/20  
Arizona Department of Human Services \$50,000  
Traumatic Brain Injury, Alzheimer's Disease, Probiotics, and Cognitive Function

## **Pending Grants**

Diego Mastroeni (Co-I)  
(Sierks, Michael) 7/1/2019-6/30/2024  
HHS: National Institutes of Health (NIH) \$1,203,175  
Characterization of toxic LBD-related protein variants using highly selective disease-specific reagents

Diego Mastroeni (PI) 9/1/2020-8/31/2025  
HHS: National Institutes of Health (NIH) \$469,918  
An Epigenetics Perspective on the Divergence from Aging to Alzheimer's disease

Diego Mastroeni (PI) 7/1/2020-6/30/2023  
Arizona State University Foundation (ASUF) \$100,000  
The Effect of ApoE-ε4 Homozygosity on Human Microglial Clearance Mechanisms

Benjamin Readhead (PI) 7/1/2020-6/30/2023  
Harvard Medical School \$89,011  
Developing aged cerebral organoids through RNA deconvolution

Benjamin Readhead (Co-I)  
(Qiu, Ji) 6/1/2020-5/31/2021  
Arizona State University Foundation (ASUF) \$100,000  
Anti-microbial immune response in Alzheimer's disease

Benjamin Readhead (Co-I)  
(Mastroeni, Diego) 12/1/2020-11/30/2022  
HHS: National Institutes of Health (NIH) \$150,000

Multi-Omic, multi organ approach to address a causal mechanism for cholinergic dysfunction in Alzheimer's disease

Benjamin Readhead (Co-I)  
(Mastroeni, Diego) 12/1/2020-11/30/2022  
HHS: National Institutes of Health (NIH) \$188,102  
Microglial Peripheral Exosomes Predict CNS Microglial Activation State

Benjamin Readhead (Co-I)  
(LaBaer, Joshua) 12/1/2020-11/30/2025  
HHS: National Institutes of Health (NIH) \$534,622  
Exploiting anti-microbial immune response for lung cancer diagnosis

Sarah Stabenfeldt (MPI)  
HHS-NIH-NINDS Lifshitz/Stabenfeldt (MPI) 07/01/20-06/30/25  
Precision targeting of the rod microglia variant in neurological disease

Sarah Stabenfeldt (Co-I)  
DOD-DARPA/BOT Abbas (PI) 10/01/20-09/30/25  
Personalized Neurotechnologies to Bridge the Gap: Promoting Recovery, Health and Function after SCI (PN-BG+)

Sarah Stabenfeldt (MPI)  
HHS-NIH-NINDS Stabenfeldt/Sirianni (MPI) 10/01/20-9/30/25  
Exploiting sex-dependent brain injury response for nanoparticle therapeutics

Sarah Stabenfeldt (MPI)  
HHS-NIH-NINDS Stabenfeldt/Holloway/Newbern (MPI) 10/01/20-9/30/25  
Leveraging SDF-1a signaling to modulate interneuron transplant and recovery after TBI

Sarah Stabenfeldt (Co-PI)  
NSF Bennett (PI) 09/01/20-08/31/23  
From Ethics Training to Ethical Formation: Developing and Testing a New Model for Responsible Research in STEM

Yalin Wang (PI) 7/1/2020-6/30/2022  
HHS: National Institutes of Health (NIH) \$157,771  
Developing a Univariate Neurodegeneration Imaging Biomarker with Optimal Transportation

Yalin Wang (PI) 9/1/2020-8/31/2025  
HHS: National Institutes of Health (NIH) \$306,048  
Hierarchical Bayesian Analysis of Retinotopic Maps of the Human Visual Cortex with Conformal Geometry

Yalin Wang (PI) 9/1/2020-8/31/2022  
University of California: San Diego \$21,369  
Cerebellar morphometry in the blind via tensor-valued and random field statistics

Yalin Wang (PI) 9/1/2020-8/31/2021  
Children's Hospital Los Angeles \$99,997  
Influence of APOE4 Genotype on Neonatal Cortical Morphology

Melissa Wilson (PI) NIH R21 New approaches to accounting for sex as a biological variable in Alzheimer's Disease	07/2020 – 06/2022 \$275,000
Eric Reiman (PI) 1 R01 AG069453-01 (Reiman/Su/Chen/Langbaum/Caselli) NIH/NIA APOE in the Predisposition to, Protection from and Prevention of Alzheimer's Disease	7/01/2020-6/31/2025 \$3,929,639 Annual DC
Eric Reiman (Co-I) NIH R01 NIH/NIA via University of Wisconsin The Neighborhoods Study: Contextual Disadvantage and Alzheimer's Disease and Related Dementias (ADRD)	12/1/2020-11/30/2025 \$58,083 Annual DC
Yi Su (Site PI) 1R01AG061122-01 (Didsbury) T3D Therapeutics/NIH/NIA Phase 2 Randomized, Double-Blind, Placebo-Controlled Clinical Trial of T3D-959 in Mild to Moderate Alzheimer's Disease Subjects	3/1/2020-2/28/2022 \$14,256 Annual Direct Costs
Yi Su (MPI) NIH R01 PAR-17-031 (Chang) University of Arizona/NIH/NIA Predictive Network Study of Blood-based Aging-related Metabolic Biomarker and Therapeutics for Preclinical Stage of Alzheimer's Disease	9/1/2020-8/31/2022 \$93,131 Annual DC
Yi Su (Co-I) NIH R21 (Pan) Arizona State University/NIH/NIA Deep Learning Algorithms for Brain Image Classification	4/1/2020-3/31/2023 \$30,019 Annual DC
Yi Su (PI) 1 R01 AG069453-01 (Reiman/Su/Chen/Langbaum/Caselli) NIH/NIA APOE in the Predisposition to, Protection from and Prevention of Alzheimer's Disease	7/01/2020-6/31/2025 \$3,929,639 Annual DC
Yi Su(Co-I) NIH R41/R42 TBD (Lure) NIH STTR via MS Technologies Multi-Modality Image Data Fusion and Machine Learning Approaches for Personalized Diagnostics and Prognostics of MCI due to AD	1/1/2021-12/31/2022 \$241,309
Yi Su (Co-I) NIH R01 TBD (Wang) NIH/NIA via ASU Multivariate morphometry statistics and sparse dictionary learning for longitudinal structural MRI analysis in nondemented individuals	7/1/2020-6/30/2024 \$987,870
Yi Su (Co-I) NIH R21 TBD (Wang)	7/1/2020-6/30/2022

NIH/NIA via ASU	\$17,230
Developing a Univariate Neurodegeneration Imaging Biomarker with Optimal Transportation	
Yi Su (Co-I)	
NIH R01 TBD (Zhou)	7/1/2020-6/30/2025
NIH via WUSTL	\$311,873
Precision PET parametric imaging of APOE $\epsilon$ 4 effects on brain tau deposition	
Kewei Chen (PI)	
1 R01 AG069453-01 (Reiman/Su/Chen/Langbaum/Caselli)	7/01/2020-6/31/2025
NIH/NIA	\$3,929,639 Annual DC
APOE in the Predisposition to, Protection from and Prevention of Alzheimer's Disease	
Kewei Chen (Co-I)	
NIH R01	12/1/2020-11/30/2025
NIH/NIA via University of Wisconsin	\$58,083 Annual DC
The Neighborhoods Study: Contextual Disadvantage and Alzheimer's Disease and Related Dementias (ADRD)	
Jessica Langbaum (PI)	
1 R01 AG069453-01 (Reiman/Su/Chen/Langbaum/Caselli)	7/01/2020-6/31/2025
NIH/NIA	\$3,929,639 Annual DC
APOE in the Predisposition to, Protection from and Prevention of Alzheimer's Disease	
Jessica Langbaum (PI)	
1 R33 PAR-18-877	9/01/2020-8/31/2025
NIH	\$799,998.00 Annual DC
Optimizing Research Infrastructure of Registries to Accelerate Participant Recruitment into Alzheimer's Focused Studies	
Michael Malek-Ahmadi (PI)	
ADCC Pilot Project Grant	7/1/2020-6/30/2021
NIH via ADCC	\$37,221
Cardiovascular Genotype and APOE $\epsilon$ 4 Carrier Status Interaction Effects on Amyloid Load in Pre-Clinical Alzheimer's Disease.	
Thomas Beach	7/1/19-6/30/20
NIH R01 via Michigan State University	\$17,000 Annual DC
Targeting raphe-striatal neuroplasticity in L-DOPA-induced dyskinesia	
Thomas Beach	2/1/19-1/31/21
NIH U01 via Case University	\$22,014 Annual DC
Assessing skin biomarkers for preclinical diagnosis of PD and non-PD Parkinsonism	
Thomas Beach	4/1/19-3/31/20
NIH R01 via ASU	\$6,994 Annual DC
Structural characterization of toxic oligomeric beta-amyloid aggregates	
Thomas Beach	9/1/18-8/31/23
NIH U01 via ASU	\$15,000 Annual DC
Center for Neurodegenerative Structural Discovery	

Thomas Beach NIH U54 via ASU Characterization of toxic-LBD-related protein variants using highly Selective disease-specific reagents	7/1/19-6/30/24 \$10,000 Annual DC
Thomas Beach AHA via Carl T. Hayden Medical Research Foundation Medin-induced vasculopathy: a new paradigm in aging-associated vascular dementia	7/1/19-6/30/21 \$7,383 Annual DC
Thomas Beach NIH R01 via ASU Oligomeric variants of alpha synuclein trigger microglial activation in Parkinson's disease	9/1/19-8/31/24 \$60,000 Annual DC
Thomas Beach NIH R01 via Mayo-Jacksonville Identifying functional mutations and aberrant RNA isoforms in top Alzheimer's disease genes using long-read sequencing in brain tissue	9/1/19-8/31/24 \$25,000 Annual DC
Thomas Beach MJFF via Mount Sinai Screening & quantification of peripheral $\alpha$ -synuclein pathology using artificial intelligence	10/15/19-10/14/20 \$17,762 Annual DC
Thomas Beach NIH R01 via Iowa State Progression of Rationale Based Progression Biomarkers for Parkinson's disease dementia	9/1/19-8/31/24 \$25,000 Annual DC
Thomas Beach NIH R01 via ASU Conformational protein variants as biomarkers for cognitive decline in Parkinson's disease	9/1/19-8/31/24 \$44,191 Annual DC
Thomas Beach NIH R01 via Mayo Physiological EEG Progression Biomarkers for Cognitive Impairment in Parkinson's disease dementia	9/1/19-8/31/24 \$11,101 Annual DC
Thomas Beach AZDHS via ABRC via Mayo Submandibular gland needle core biopsy as a tissue biomarker for the diagnosis of Parkinson's disease and the monitoring of disease progression	7/1/19-6/30/22 \$100,028 Annual DC
Thomas Beach NIH R01 via TGen Extracellular RNA Biomarkers of Parkinson's Disease Dementia	9/1/19-8/31/24 \$132,999 Annual DC
Thomas Beach NIH R01 via Ohio State University The role of ectodermal-neural cortex 1 in selective vulnerability in aging and Alzheimer's disease	4/1/20-3/31/25 \$45,230 Annual DC

Thomas Beach NIH R01 via Case Western University Skin biomarkers for diagnosing and characterizing AD and ADRD	4/1/20-3/31/25 \$74,953 Annual DC
Thomas Beach NIH DP2 via University of Hawaii Using genetic instruments for prostate tissue specific DNA methylation and protein biomarkers identification and risk prediction for prostate cancer	9/30/20-9/29/24 \$19,014 Annual DC
Thomas Beach1 European Union via Maastricht University (Netherlands) via Arizona State University DCBrightlands e-infrastructure for Neurohealth (BReIN)	0/1/19-9/30/20 \$100,267 Annual
Thomas Beach NIH R01 via Mayo Clinic – Jacksonville Using long-range technologies as a multi-omic approach to understand Alzheimer’s disease in brain tissue	7/1/20-6/30/25 \$25,000 Annual DC
Thomas Beach NIH U01 via Case University Assessing skin biomarkers for preclinical diagnose of PD and non-PD Parkinsonism	2/1/19-1/31/21 \$21,000 Annual DC
Thomas Beach NIH R01 via Iowa State Evaluation of Rationale-Based Progression Markers for Parkinson’s Disease Dementia	7/1/20-6/30/25 \$110,000 Annual DC
Thomas Beach NIH R01 via TGen Extracellular RNA Biomarkers of Parkinson’s Disease Dementia	7/1/20-6/30/25 \$22,000 Annual DC
Thomas Beach NIH R01 via University of California – San Diego In situ astrocyte to neuron trans-differentiation in mouse models of Alzheimer’s disease and Parkinson’s disease	7/1/20-6/30/25 \$18,000 Annual DC
Thomas Beach NIH R01 via University of Alabama – Birmingham Synapse loss is an early pathological event in Lewy body dementias	7/1/20-6/30/25 \$5,000 Annual DC
Thomas Beach NIH R01 via University of California – Santa Barbara The complex interaction between Alzheimer’s drivers and aging	7/1/20-6/30/25 \$55,000 Annual DC
Thomas Beach NIH R01 via Arizona State University 3-D conformation of key toxic beta-amyloid and tau variants in AD	7/1/20-6/30/25 \$11,000 Annual DC
Thomas Beach NIH R01 Blinded Comparison of Different Alpha-Synuclein Seedling Assays as	7/1/20-6/30/24 \$690,581 Annual DC

## Cutaneous Biomarkers of Lewy Body Dementia

Geidy Serrano 07/1/20-6/30/24  
NIH R01 \$690,581 Annual DC  
Blinded Comparison of Different Alpha-Synuclein Seedling Assays as Cutaneous Biomarkers of Lewy Body Dementia

Rita Sattler (PI) 07/01/20-06/30/25  
NIH/NINDS RO1 (Sattler/Van Keuren-Jensen/Zarnescu)  
Mechanisms of TDP-43 proteinopathies in common dementias

Rita Sattler (PI) 07/01/20-06/30/22  
TARGET ALS ((Tardiff (Yumanity Inc)/Sattler/Zarnescu)  
Validation of Cyp51 as a therapeutic target for ALS/FTLD

Rita Sattler (PI) 07/01/20-06/30/22  
TARGET ALS (Alworth (Acurastem Inc)/Ichida/Sattler/Gao)  
Validation of Cyp51 as a therapeutic target for ALS/FTLD

Rita Sattler (PI) 07/01/20-06/30/22  
VALLEY RESEARCH PROPOSAL (Gustin/Sattler)  
Interplay between C9orf72 ALS and neurotropic viruses: impact on viral and ALS disease pathogenesis

Rita Sattler (Mentor) 07/01/20-06/30/21  
BNI FOUNDATION (Gittings)  
Identifying specific cellular and molecular signatures that influence the development of cognitive impairment in C9orf72 FTD/ALS using single cell transcriptomics and patient-derived neuronal cell culture models

Rita Sattler (Mentor) 07/01/20-06/30/22  
EMBO POSTDOCTORAL FELLOWSHIP AWARD (Gittings)  
Identifying specific cellular and molecular signatures that influence the development of cognitive impairment in C9orf72 FTD/ALS using single cell transcriptomics and patient-derived induced pluripotent stem cell culture models

Ashley Stokes 04/01/20-03/31/25  
UG3CA247606-01 (Quarles)  
NIH/NCI  
Structural and Functional Imaging for Therapy Response Assessment in Brain Cancer

Layla Al-Nakkash and Minsub Shim (PIs) 07/01/21-06/30/21  
U.S. National Academy of Medicine \$50,000  
Genistein and Exercise Ameliorate Senescence and Inflammation to Preclude Progression of Alzheimer's Pathology.

Mitra Esfandiarei (Co-I) 07/01/20-06/30/24  
NIH R15 \$450,000  
Identifying the role of maternal and postnatal dietary deficiencies on peripheral and cerebral vasculature and stroke outcome in middle- and old-age offspring.

Nafisa Jadavji (PI) American Federation for Aging Research 2020 Glenn Foundation for Medical Research and AFAR Grants for Junior Faculty LOI Identification of developmental factors involved in neurodegeneration outcomes during old age	01/08/20-01/07/21 \$100,000
Nafisa Jadavji (PI) NIH R15 REAP Identifying the role of maternal and postnatal dietary deficiencies on peripheral and cerebral vasculature and stroke outcome in middle- and old-age offspring	01/01/21-12/31/23 \$450,000
Garilyn Jentarra, (Co-PI) & Haiwei Gu (PI) Arizona State University, College of Health Solutions, Jumpstart Targeting Fatty Acid Metabolism in Alzheimer's Disease: A Special Interest in Lauric acid	05/01/20-04/28/21 \$18,000
Jose Hernandez (Co-PI) Arizona Biomedical Research Center Elucidating the role of <i>Rhipicephalus sanguineus</i> (the Brown dog tick) as a vector for Rocky Mountain Spotted Fever (RMSF) transmission in Arizona	04/01/20-04/01/23 \$446,495
Vanthida Huang (PI) AFPE In vitro Activity of Ceftaroline Alone and in Combination with Daptomycin at High and Low Doses Against Methicillin-resistant Staphylococcus aureus in an in vitro Pharmacodynamic Model	06/01/20-05/31/21 \$5000
Kathy Lawson (PI) NIH R21 Oncogenic P-cadherin Signaling in Oral Preneoplasia	04/01/20-03/31/22 \$412,500
Ashlesh Murthy (PI) NIH R01 Synthesis of Clp Protease Proteolytic Subunit (ClpP) Activators to Treat Chlamydial Infections	10/01/20-09/30/25 \$210,445
Mark Olsen (PI) Winn Feline Foundation Developing a Safe and Effective Anticoronaviral Therapy for Client-owned Cats with FIP	04/01/20-12/31/21 \$7,000
Ann Revill (PI) NIH R15 Cholinergic modulation of XII motoneurons and premotoneurons	07/01/20-06/30/23 \$449,999
Ann Revill (Co-Investigator) NIH R15 <i>In vitro</i> and <i>in vivo</i> analysis of metabolic adaptations in non-small cell lung cancer as a function of p53 and energy status using a novel, direct ATP chelation methodology	07/01/20-06/30/23 \$300,000
Ann Revill (PI) Sleep Research Society Foundation Career Development Award Excitatory and inhibitory cholinergic modulation of XII motoneurons across postnatal maturation	07/01/20-06/30/21 \$50,000

Ann Reville (PI) McKnight Endowment Fund for Neuroscience Costs McKnight Scholar Award Excitatory and inhibitory cholinergic modulation of XII motoneurons and XII premotoneurons across postnatal development	07/01/20-06/30/23 \$225,000
Emily Cope (PI), J Gregory Caporaso (PI), Schwartz (Investigator) PAR-19-071 NIH Toward a mechanistic link between Alzheimer's Disease and the gut microbiome using quantitative Stable Isotope Probing	10/01/20-09/30/22 \$412,414
J Gregory Caporaso (PI) PAR-15-331 NIH Advancing our Understanding of Cancer and the Human Microbiome with QIIME 2	04/01/20-03/31/25 \$707,445
J Gregory Caporaso (Co-I) PA-18-876 (Duca) NIH Oligofructose restores nutrient-sensing mechanisms regulating glucose production to improve glucose homeostasis via alterations in small intestinal microbiota	09/01/22-08/31/24 \$45,427
J Gregory Caporaso (Investigator) RFA-CA-19-039 (Crandall/Johnson) NIH MetaScope - A unified bioinformatics framework for microbiome sequencing studies in cancer	04/01/20-03/31/23 \$24,999
J Gregory Caporaso (Investigator) PAR-18-654 (Herbst-Kralovetz) NIH Longitudinal multi-omics analysis and elucidation of the functional impact of the cervicovaginal microenvironment in Hispanic women to reduce cervical cancer disparities	04/01/20-03/31/25 \$21,837
J Gregory Caporaso (Investigator) PAR-18-714 (Pearson) NIH Are Minority Health Disparities in MRSA/MSSA Infections Related to Carriage and Social Relationships?	09/01/20-08/31/23 \$148,264
J Gregory Caporaso (Investigator) NSF 20-513 (Marks) National Science Foundation Discovering in reverse: using isotopic translation of omics to reveal ecological interactions in microbiomes	10/01/20-09/30/25 \$358,141
J Gregory Caporaso (Investigator) RFA-RM-19-012 (Bokulich) NIH Integrating iHMP and metabolomics workbench data to guide microbial ecosystem analysis across the human body	09/01/20-08/31/21 \$195,490

J Gregory Caporaso (Investigator) NSF 20-508 (Luo) National Science Foundation BII-Implementation: Integration Institute of Carbon Metabolism (IICM): Molecule to Earth	10/01/20-09/30/25 \$1,789,109
J Gregory Caporaso (Investigator) PAR-19-198 (Herbst-Kralovetz) NIH Microbiome-Mediated Immune Mechanisms that Affect the Efficacy of Endometrial Cancer Therapies	07/01/20-06/30/25 \$39,115
Emily Cope (PI) PA-19-056 (Duddleston/Buck/Caporaso/Cope) NIH Toward microbial intervention for lean mass loss: the role of the gut microbiome in essential amino acid synthesis	04/01/20-03/31/25 \$152,653
Emily Cope (PI) BPM000338 Arizona Biomedical Research Commission (ABRC) Human genetic variation and the sinonasal microbiome in chronic rhinosinusitis	07/01/19-06/30/22 \$225,000
Emily Cope (PI) R15 NIH/NIAID Defining the role of the sinonasal mucosal mycobiome in chronic rhinosinusitis	07/01/19-06/30/21 \$471,963
Richard Caselli (PI) R01AG031581 (Reiman/Caselli) APOE in the Predisposition to, Protection from and Prevention of Alzheimer's Disease	07/01/20-06/30/25
Dona Locke (Co-I) R01AG031581 (Reiman/Caselli) APOE in the Predisposition to, Protection from and Prevention of Alzheimer's Disease	07/01/20-06/30/25
Matthew Huentelman (Co-I) R56AG045571 (Rogalaski) NIH (Northwestern University) Exceptional Cognitive Aging: Neuropsychologic, Anatomic and Pathologic	05/01/19-04/30/24 \$55,000
Matthew Huentelman (Co-I) R01AG059627 (Caccamo) NIH Identify common mechanisms of neurodegeneration between Alzheimer's disease and Down syndrome	09/01/19 -08/31/20 \$54,808
Matthew Huentelman (Co-I) R01 NIH (Gallitano) Mechanism of 5HT2AR Regulation by Egr3	09/01/23-08/31/24 \$13,342

Matthew Huentelman (Co-PI) DARPA (Broadrick) HR001119S0021-MBA-FP-002 (BTO) Peerless Operator Biologic Aptitude (Peerless)	10/01/19-03/31/21 \$476,774
Matthew Huentelman (Co-I) R01 (Rogalaski) NIH Cognitive SuperAging: A model to explore resilience and resistance to aging and Alzheimer's disease	04/01/20-03/31/25 \$40,879
Matthew Huentelman (Co-I) R01 (Barnes) NIH NPTX2: Preserving memory circuits in normative aging and Alzheimer's Disease	07/01/20-06/30/25 \$126,560
Matthew Huentelman (Co-PI) Grant (Koshy) Chan Zuckerberg Leveraging Toxoplasma gondii to understand CNS-immune system	03/01/20-02/28/22 \$76,087
Matthew Huentelman (Co-I) R21 (Grilli) NIH Tracking autobiographical thoughts: a smartphone-based approach to the detection of cognitive and neural markers of Alzheimer's disease risk	07/01/20-06/30/25 \$16,500
Matthew Huentelman (Co-I) R01 (Hale) NIH Targeting Resident Cardiac Fibroblast Subpopulations for Protection Against Fibrosis	07/01/20 – 06/30/25 \$80,850
Matthew Huentelman (Co-I) U01 (Olive) NIH Genetics and Epigenetics of Heroin Demand in the Sprague-Dawley Rat	07/01/20-06/30/25 \$932,343
Matthew Huentelman (Co-I) UC2 (Gray) NIH Patient engagement and genomic analysis in early-onset colorectal cancer	07/01/20-06/30/25 \$30,960
Matthew Huentelman (PI) R21 NIH Deployment of a wearable device for near real-time reactive phenotyping and biosampling	07/01/20-06/30/22 \$175,000
Matthew Huentelman (Co-I) R01 (Reiman) NIH APOE in the Predisposition to, Protection from and Prevention of Alzheimer's Disease	07/01/20-06/30/25 \$74,537

Matthew Huentelman (Co-I) R01 (Geula) NIH	06/01/20-05/31/25 \$57,607
Molecular, Cellular, and Clinicopathologic Signatures of Co-morbid TDP-43 in Alzheimer's Disease	
Winnie Liang (Co-I) U01 (Weitzel) NIH	09/01/19-08/31/23 \$95,000
Enhancing Precision Prevention through Family Informative Genetic Testing (FIT)	
Winnie Liang (PI) R21 NIH	09/01/20-08/31/22 \$69,044
Evaluation of the impact of acupuncture on mild cognitive impairment patients	
Winnie Liang (Co-I) U2C (Willman) NIH	07/01/20-06/30/25 \$808,527
Engagement of American Indians of Southwestern Indigenous Tribal Nations in Cancer Genome Sequencing-Genomic Characterization Unit	
Kendall Van Keuren-Jensen (Consortium PI) UG3UH3 (Wong) NIH	07/01/20-06/30/23 \$145,000
Acoustofluidic (AF) Separation, Purification and Raman Spectral Fingerprinting of Single EVs: From Cell of Origin to Target Cell and Biofluids	
Kendall Van Keuren-Jensen (Co-I) Grant (Bowser) Muscular Dystrophy Association	08/01/19-07/31/21 \$17,673
Novel Mouse Models of ALS and Myopathy-linked Matrin 3 Mutations	
Kendall Van Keuren-Jensen (Co-I) R01 supplement (Zarnescu) NIH	06/01/19-05/31/20 \$30,000
RNA dysregulation in neurodegeneration	
Kendall Van Keuren-Jensen (PI) R01 NIH	09/01/19-08/31/24 \$352,458
Extracellular RNA biomarkers of Parkinsons disease dementia	
Kendall Van Keuren-Jensen (Co-I) R61/R33 (Wong) NIH	07/01/19-06/30/24 \$13,937
Development of Salivary exRNA Biomarkers for Chronic Neck Pain and Endpoint	
Kendall Van Keuren-Jensen (Co-I) R21 NIH	04/01/20-03/31/22 \$29,160

Novel knock-in mouse models of ALS and myopathy-linked Matrin 3 mutations

Alexander, Gene (PI)  
(multi PI: Raichlen; co-I's: Trouard, Hishaw, Simpson) 7/1/20 – 6/30/25  
NIA R01AG067200 \$3,837,378 Total Costs  
Physical Activity Predictors of Cognitive and Brain Health in the Risk for Alzheimer's disease

Alexander, Gene 9/1/19 – 8/31/24  
McKnight Brain Research Foundation \$60,000 Total Costs  
Transcutaneous Vagal Nerve Stimulation and Cognitive Training to Enhance Cognitive Performance in Healthy Older Adults

Barnes Carol (PI)  
co-I's: Albert, Bilgin, Brinton, N-K Chen, Z Chen 2021 - 2026  
Coon, Doyle, Duarte, Ellingson, Hay, Huentelman Lafleur, Levin \$55,564,780 Total Costs  
Mehl, Merchant, Padi, Resnick, Rodgers, Rundek, Runyon, Ryan,  
Schork, Sternberg, Trouard, Worley)  
NIH/NIA U19 AG065169  
Precision Aging Network: Closing the Gap Between Cognitive Healthspan and Human Lifespan

Barnes, Carol (PI) 07/01/20 – 06/30/25  
(co-I's: Huentelman, Trouard, Worley) \$6,995,440 Total Costs  
NIH NIA RO1 AG062652  
NPTX2: Preserving memory circuits in normative aging and Alzheimer's disease

Cai, Minying (PI; Co-I: Barnes) 09/01/20 – 08/31/22  
NIH \$417,206 Total Costs  
Study of Melanocortin Receptors Expression Correlated with Spatial Memory

Chen, Nan-kuei (UA PI; Stokes PI) 09/01/20 – 08/31/25  
NIH Sub (St Joseph's Hospital and Medical Center) \$187,151 Total Costs  
Multi-Echo & Multi-Contrast Functional MRI Using Combined Spin and Gradient-Echo (SAGE) Methods

Chou, Ying-Hui (PI) 05/01/20 – 04/30/25  
co-I's Alexander, Barnes, Bedrick, Mohler, Rapcsak, Ryan) \$3,629,832 Total Cost  
NIH R01 AG062543  
Enhancement of Hippocampal Plasticity Using Repetitive Transcranial Magnetic Stimulation

Chou, Ying-Hui (PI) 08/01/20 – 07/31/25  
co-I's Alexander, Bedrick, Chen, Fisher, Rapcsak, Ryan) \$3,546,143 Total Cost  
NIH R01  
Accelerated Theta Burst Stimulation for Individuals at a High Risk of Developing Alzheimer's Disease

Ekstrom, Arne (PI; co-I's: Andrews-Hanna, Drake, Grilli) 07/01/20 – 06/30/25  
NIH NINDS NS109819 \$3,503,611 Total Costs  
Precision and binding as two dimensions of medial temporal lobe amnesia

Fernandez (PI; co-I's: Allen, Grandner, Kilgore, Wilson) 08/01/20 – 07/31/25  
NIH

Nocturnal wakefulness and suicidality: neurobiological and neuropsychological concomitants in young adults Gaffney, Keven (PI) NIH R43 AG067884	04/01/20 – 03/31/21 \$450,000 Total Costs
Optimizing of Small Molecule Lipoxin Mimetics for the Treatment of Alzheimer's Disease	
Grilli, Matthew (PI; co-I's: Andrews-Hanna, Mehl) NIH/NIA R01 AG068098	07/01/20 – 06/30/25 \$2,074,848 Total Costs
Tracking autobiographical thoughts: a smartphone-based approach to the detection of cognitive and neural markers of Alzheimer's disease risk	
Khanna, May (PI; co-I: Wang) Alzheimer's Drug Discovery Foundation	09/01/19 – 08/31/21 \$600,000 Total Costs
Targeting Necroptosis for Neurodegenerative Disease Therapy	
Khanna, May (PI; co-I's: Roveda, Churko) NIH	07/01/20 – 06/30/25 \$4,020,641 Total Costs
Leveraging Chemical Biology to Study Neurodegenerative Diseases	
Khanna, May (PI) National Ataxia Foundation	03/01/20 – 02/28/21 \$100,000 Total Costs
Developing phenotypic screens for FDA approved drugs that can be neuroprotective in PCH1B	
Khanna, May (PI; co-I's: Churko, Wang) Regenerex LLC	07/20/20 – 06/30/21 \$269,777 Total Costs
Targeting Necroptosis for Neurodegenerative Disease Therapy	
Khanna, May (PI) Arizona Alzheimer's Consortium	07/01/29 – 06/30/20 \$26,050 Total Costs
Developing Inhibitors of TDP-43/Tau interaction for AD Therapeutics	
Rodgers, Kathleen (PI; co-I: Gaffney, Milnes, Vanderah ) Southern Arizona VA Health Care System	04/01/20 – 03/31/24 \$509,830 Total Costs
Small Molecule Mas Agonists for the Treating Traumatic Brain Injury	
Rodgers, Kathleen (PI) (co-I: Brinton, Gaffney) NIH	04/01/20 – 03/31/24 \$6,108,279 Total Costs
IND Enabling Studies for RASRx 1902, a novel Mas receptor agonist, for treatment of cognitive impairment in patients at risk for Alzheimer's disease	
Ryan, Lee (multi-PI) (PI's: Lazar; Sweitzer; co-I's: Hay, Konhilas, Bedrick) NIH R11	07/01/20 – 06/30/25 \$8,696,506 Total Costs
Safety and Efficacy of Angiotensin1-7 for cognitive impairment in heart failure patients at risk for vascular dementia and Alzheimer's disease and related dementias	
Saranathan, Manoj (PI) co-Is: Chen, Rapcsak, Guzman, Chou, Grilli, Pattersen, Vedantham) NIH NIA/NIBIB 10028636 ID	07/01/20 – 06/30/25 \$1,760,931 Total DC

High Resolution Anatomical and Structural-Functional Connectivity Imaging of Thalamic Nuclei in Alzheimer's Disease

Su, Judith (UA PI) California Institute of Technology (NIH Prime) Prediction of 3D Structure, Ligand Binding, G-Protein Signaling, and beta-arrestin Signaling of all 25 Human Bitter Taste Receptors with Experimental Validation	12/01/20 – 11/30/25 \$351,815 Total Costs
Su, Judith (PI) Partnership for Clean Competition Ultra-Sensitive Detection of Multiple Performance Enhancing Drugs Using Optical Resonators	07/01/20 – 06/30/23 \$400,000 Total Costs
Su, Judith (PI) NIH Label-free Ultra-Sensitive Biomolecular Detection for Basic Science and Translational Medicine	07/01/20 – 06/30/25 \$1,822,950 Total Costs
Su, Judith (PI) Alfred P Sloan Foundation Label-free Single-Molecule Detection for Basic Science and Translational Medicine	09/15/20 – 09/14/22 \$75,000 Total Costs
Toosizadeh, Nima (PI) co-Is: Laksari, Mohler, Parthasarathy, Rapcsak Vemulapalli, Wendel NIH/NIA AG063738 Predicting Longitudinal Health Outcomes in Hospitalized Older Adults with Multiple Chronic Conditions Including Alzheimer's and Related Dementias Using a Combined Physical and Cognitive Function Tool	04/02/20 – 03/31/24 \$1,819,480 Total Costs
Trouard, Ted (PI) Microvascular Therapeutics, LLC BIR Phase I: Novel Theragnostic Application of Nano Contrast Agents for Early Detection and Treatment of Alzheimer's Disease	04/06/20 – 04/05/21 \$109,110 Total Costs
Weinkauf, Craig (PI; co-I: Alexander) NIA RO1 Extracranial Carotid Atherosclerosis Contributions to Cognitive Impairment and Alzheimer's Disease Risk	09/01/20 – 08/31/25 \$4,909,773 Total Costs
Weinkauf, Craig (PI) CVPath Institute Role of CD163 Macrophages in Atherosclerosis Progression	12/01/20 – 11/30/25 \$86,075 Total Costs
Weinkauf, Craig (PI) Arizona Alzheimer's Consortium Extracranial Carotid Atherosclerosis Contributions to Alzheimer's Disease Risk	07/01/20 – 06/30/21 \$45,927 Total Costs
Witte, Russell (PI) co-I's: Becker, Chen, Chou, Cowen, Gothard, Weinberg) NIH/NIBIB U01 EB028662 4D Transcranial Acoustoelectric Imaging for High Resolution Functional Mapping of Neuronal Currents	9/30/2019 – 9/29/2020 \$303,425 Annual DC

Yin, Fei (co-I; PI: Hammer) American Epilepsy Society Testing the Role of Early Mitochondrial Dysfunction in SCN8A Epilepsy	06/10/19 – 06/09/20 \$15,000 Total Costs
Zhou, Wei (PI: co-I's: Becker, Hsu, Saranathan, Trouard) NIH/NINDS Long-term Cognitive Effects of Subclinical Microembolization Associated with Carotid Interventions	07/01/20 – 06/30/25 \$2,210,271 Total Costs
Zhou, Wei (PI: co-I's: Becker, Hsu, Saranathan, Trouard) NIH Cognitive Effects of Carotid Disease and Carotid Interventions in Dementia	09/01/20 – 08/31/25 \$2,847,009 Total Costs
Jonathan Lifshitz (Co-PI) GRANT12660842 (Handmaker) Office for Victims of Crime Maricopa County Collaboration on Concussion from Domestic Violence (MC3DV)	10/01/18-09/30/20 \$879,944
Jonathan Lifshitz (PI) R21 NS111326 (Law) National Institutes of Health Effectiveness of Cognitive Rehabilitation Depends on Hippocampal Plasticity Following Brain Injury	04/01/19-03/31/21 \$422,125
Jonathan Lifshitz (Co-I) R21 NS112943 (Stabenfeldt) Arizona State University   NIH Sex Dependent Considerations for TBI Nanotherapeutics	07/01/19-06/30/24 \$1,172,399
Jonathan Lifshitz (Co-I) R01 NS111228 (Rowe) National Institutes of Health Extinguishing the Fire: Eliminating Microglia to Attenuate Inflammation-Induced Sleep	09/01/19-03/31/24 \$2,498,430
Jonathan Lifshitz (PI) Grant (Stabenfeldt) Chan Zuckerberg Initiative Precision Targeting of the Rod Microglia Variant in Neurological Disease (Single-Cell Analysis of Inflammation)	03/01/20-02/28/22 \$350,000
Jonathan Lifshitz (PI) R01 NS110795 (Subbian) National Institutes of Health Analytical Modeling of Acquired Neurological Injury with Rich Experimental Data Sets	04/01/20-03/31/25 \$3,641,681
Jonathan Lifshitz (Mentor) F31 NS113408 (Giordano) NINDS National Institutes of Health Precision Identification and Targeting of Rod Microglia in Diffuse Brain-Injured Cortex Time	04/01/20-03/31/25 \$227,240

Jonathan Lifshitz (Co-PI) I01 _____ (Migrino) U.S. Dept. of Veterans Affairs Mechanistic Role of Vascular Dysfunction in TBI-Mediated Cognitive Dysfunction	04/01/20-03/31/24 \$1,200,000
Jonathan Lifshitz (Co-I) R01 NS116012 (Rowe) National Institutes of Health Microglia in Sleep	04/01/20-03/31/25 \$2,486,468
Jonathan Lifshitz (PI) Grant (Law) Phoenix Children's Hospital Virtual Cognitive Rehabilitation	06/01/20-05/31/21 \$150,000
Jonathan Lifshitz (PI) Grant (Oatman/Sukhina) Phoenix Children's Hospital Pediatric Traumatic Brain Injury and Hypothalamic-Pituitary Disorders in Arizona: A Prospective Pilot Study	06/01/20-05/31/21 \$150,000
Jonathan Lifshitz (PI) R01 NS118853 (Stabenfeldt) National Institutes of Health Precision Targeting of the Rod Microglia Variant in Neurological Disease	07/01/20-06/30/25 \$3,910,653
Jonathan Lifshitz (Co-I) R01 NS117919 (Rowe) National Institutes of Health Microglia as Mediators of Brain Injury-Induced Sleep Disturbances	07/01/20-06/30/25 \$2,358,482
Jonathan Lifshitz (Co-I) R13 NS _____ (Giza) National Institutes of Health Western Neurotrauma Symposium	07/01/20-06/30/21 \$30,000
Jonathan Lifshitz (Consultant) R21 NS _____ (Murray) National Institutes of Health Preclinical Evaluation Of Combination Therapy of Rolipram and Minocycline for Arresting Secondary Injury Cascade after Traumatic Brain Injury	07/01/20-06/30/22 \$275,000
Jonathan Lifshitz (Site PI) Grant (Sierks) Arizona State University   NIH Toxic Protein Variants Role in Increased Risk of ADRDs After TBI	07/01/20-06/30/25 \$1,005,905
Jonathan Lifshitz (PI) Grant Dept. of the Army – USAMRAA	10/01/20-09/30/22 \$1,906,487

Clearing Acute Lung Injury Induced by Brain Injury with Remote Ischemic Conditioning (CALIBRIC)



**Arizona Alzheimer's Consortium  
22<sup>nd</sup> Annual Scientific Conference**

**Scientific Abstracts**

**STUDYING PROTEIN INTERACTIONS AMONG THE TELOMERE PROTEIN TERF2IP, THE EPSILON ISOFORM OF GLIAL FIBRILLARY ACIDIC PROTEIN AND THE ALZHEIMER'S DISEASE ASSOCIATED PRESENILIN 1.** Abel KN, Chow AY, Gusloff R, Bae NS, Swanson MJ. Midwestern University; Arizona Alzheimer's Consortium.

Background: The ends of linear, eukaryotic chromosomes (telomeres) are protected by a complex called shelterin. Unique among its subunits is TERF2IP, which can also localize to the cytoplasm. To study cytoplasmic functions of TERF2IP, we used a yeast two-hybrid (Y2H) screen that identified a novel interaction with an isoform of glial fibrillary acidic protein, GFAP $\epsilon$ . This isoform is polymorphic, and the encoded proteins differ by a single amino acid at position 426 which can be threonine, alanine or valine. Previously, GFAP $\epsilon$  was shown to interact with the presenilin proteins, PS1 and PS2, the catalytic subunits of  $\gamma$ -secretase. Amyloid precursor protein (APP) is a substrate for secretase complexes. Sequential cleavage of APP by  $\beta$ -secretase then by  $\gamma$ -secretase produces amyloid (A $\beta$ ) peptides of varying lengths. Shorter peptides are soluble, but longer peptides aggregate to form amyloid plaques, a hallmark of the Alzheimer's disease brain. Mutations in the genes encoding APP, PS1 and PS2 lead to early onset familial Alzheimer's disease (eFAD). eFAD-associated mutations lead to increased total A $\beta$  or an increase in the ratio of the insoluble to the soluble peptides. Thus, our work is the first to show a potential link between a telomere protein and a protein involved with Alzheimer's disease.

Methods: The yeast two-hybrid (Y2H) system is a genetic method to detect protein-protein interactions. A yeast genetic screen using TERF2IP as a bait identified GFAP $\epsilon$  as an interacting protein from a human fetal brain cDNA library. Y2H studies were also done to determine the interactions among TERF2IP, GFAP $\epsilon$  and PS1. Variants of the proteins were made to determine any effects on the interactions. Additional studies are being done to determine whether the threonine allele of GFAP $\epsilon$  is phosphorylated, how such a modification may affect the interactions and what kinases may be responsible for the modification.

Results: We identified an interaction between GFAP $\epsilon$  and TERF2IP. Co-expression of PS1 resulted in a stronger TERF2IP-GFAP $\epsilon$  interaction, suggesting the interaction among the three proteins is cooperative. Different allelic variants of GFAP $\epsilon$  showed different strengths of interaction with TERF2IP. The GFAP $\epsilon$  426 threonine variant interacted most strongly with TERF2IP. This threonine is predicted to be phosphorylated by cdk5 and GSK3 kinases, both of which have been associated with Alzheimer's disease, and we are testing these kinases for activity with GFAP $\epsilon$ . Mutating position 426 to a glutamic acid to mimic phosphorylation did not change the interaction in the Y2H system. The threonine at this position is predicted to be phosphorylated in yeast, so we will test interactions between proteins produced in *E. coli*, which has no phosphorylation.

Conclusions: We identified a cooperative interaction among the telomeric protection protein TERF2IP, an isoform of GFAP and the eFAD-associated PS1. The interaction between TERF2IP and GFAP $\epsilon$  depends upon the specific GFAP $\epsilon$  variant tested, and a post-translational modification of GFAP $\epsilon$  may be important for the interaction.

**FREE-WATER ANALYSIS OF THE HIPPOCAMPAL COMPLEX IN AGING ADULTS WITH AUTISM SPECTRUM DISORDER.** Alvar J, Ofori E, Elms NE, Walsh M, Pagni B, Braden BB. Arizona State University; Arizona Alzheimer's Consortium.

**Background:** The hippocampus is a critical brain structure for memory formation and other aspects of cognition. The hippocampus and the white matter tracts connecting it to other parts of the brain are known to lose volume and integrity with aging. For populations with prior compromised hippocampal integrity, such as those with autism spectrum disorder (ASD), it is less well known how the hippocampus and its connections will respond to aging. In children with ASD, there may be an initial period of enlarged hippocampi, after which there is a trajectory of faster decline in volume compared to neurotypicals (NT). We have previously identified reduced hippocampal volumes and fornix white matter integrity in middle-age and older adults with ASD compared to matched NT adults. However, freewater (FW) may be a more sensitive structural integrity measure of the hippocampal complex, and has shown promise as a biomarker for Alzheimer's and Parkinson's disease. FW is present in the brain as cerebrospinal fluid but also accumulates within the extracellular spaces indicative of reduced gray matter density and increased axon degeneration. This study evaluated age-related hippocampal complex FW differences in adults with and without ASD across the adult lifespan. We hypothesized that adults with ASD would demonstrate a larger age association with increasing FW in the hippocampus and fornix, compared to NT adults, and that FW would be a more sensitive brain measure than traditional fractional anisotropy (FA).

**Methods:** The study consisted of 79 participants with ASD (59 male, 20 female; ages 18-70, mean=40.27 [ $\pm$ 17] years) and 77 NT participants (46 male, 31 female; ages 18-71, mean=40.33 [ $\pm$ 16] years). Hippocampal and fornix FW and FA values were generated from diffusion tensor images obtained along 32 directions using a b-value of 2500 s/mm<sup>2</sup> in the axial direction with 3 mm slice resolution. These images were then processed for eddy current, distortion, b-vec and motion correction, skull stripped, and non-linear registered using Advanced Normalization Tools (ANTs) to the subject's T1 image. FW and FA maps were calculated using custom written MatLab code and standard atlases containing the hippocampus and fornix were applied.

**Results:** The right hippocampus showed a significant diagnosis by age interaction ( $p=0.018$ ), such that the increase in FW with age was greater for adults with ASD. The left hippocampus diagnosis by age interaction approached significance ( $p=0.055$ ). Similarly, the right fornix showed a significant diagnosis by age interaction ( $p=0.044$ ), with increases in FW with age as greater for adults with ASD, and the left fornix diagnosis by age interaction approached significance ( $p=0.053$ ). FA values showed no significant diagnosis by age interactions.

**Conclusions:** In the hippocampus and fornix, the association between increasing FW and increasing age was more pronounced for adults with ASD than matched NT adults. This may mean that as adults with ASD age, these regions will degenerate faster than their NT peers, which could have implications for accelerated age-related memory decline. However, a notable limitation is the cross-sectional nature of the study. Our ongoing longitudinal study will inform a more definitive picture of brain aging with ASD.

**EXPANDING ARIZONA'S DEMENTIA-CAPABLE SYSTEM.** Angulo A, Cortés M, Carbajal B, Carbajal L, Cordova L, Weatherall Z, Gómez-Morales A, Glinka A, Rice GS, Gonzalez MS, Coon DW. Arizona State University; Arizona Alzheimer's Consortium.

Background: There are 5.8 million people living with Alzheimer's disease (AD) in the United States. It is estimated that by 2050, the number of individuals with Alzheimer's Disease will double to 13.8 million. Arizona will experience the greatest proportional increase of people living with Alzheimer's between now and 2025. Among the population affected with AD, there are three vulnerable groups where community partners can join efforts to serve the community in a more comprehensive manner. These include (a) people living alone with ADRD who may or may not have a family caregiver, (b) people with Down Syndrome or another Intellectual and Developmental Disability (DS/IDD) aging with ADRD and their family caregivers, and (c) people with ADRD and their family caregivers in the Latino community. The Administration on Community Living (ACL) estimates that approximately 14.3 million older adults are living alone. Latinos/Hispanics are 1.5 times more likely to develop Alzheimer's disease or other related dementias when compared to non-Hispanic white older adults. Individuals with DS/IDD have a genetic propensity to develop early onset ADRD, affecting between 50% and 70% of the individuals by the age 60. Dementia capable systems are designed to address the needs and concerns of all individuals, families, and communities impacted by ADRD. The overall aim of this project is to develop and expand ADRD programs and services across Maricopa/Pima counties through educational workshops, case management services, and evidence-based programs addressing the needs of these three target groups. The current presentation focuses on results from participant evaluations in the project's educational workshops to date.

Methods: Three educational presentations on ADRD and the target groups were created and delivered by community partners in English or Spanish: Living Alone with ADRD, ADRD in the Latino Community, and Dementia and DS/IDD. Presentations were offered to a variety of professionals and community members ranging from Promotoras/CHW's (Community Health Workers), case managers, to family caregivers and people with dementia. Assessment tools captured workshop attendee demographics, and information about their care recipients (if applicable) as well as their perception of benefit from the presentations provided. The assessment includes questions about meeting expectations, satisfaction, acquisition of new and useful information, and likeliness to recommend the program.

Results: A total of 59 presentations were delivered and 2,223 participants throughout Maricopa and Pima counties. Of the 1,282 participants (n) completing the tools, 64.8% self-identified as Hispanic or Latino/a, and 85.2% were females. The largest percentage of participants were case managers, care coordinators, or discharge planners (22.7%). The proportion of presentations broken down by topic were: 42.4% Latino community, 33.6% living alone, and 24.0% DS/IDD. The perception of benefit ratings were overwhelmingly positive and reflected the willingness of participants to attend other community education programs. Participants (73.3%) agreed that they felt confident that they could help these populations and 86.1% stated that they were "very likely" to recommend the project to others.

Conclusions: Data revealed that presentations were highly acceptable to participants and contributed to increased awareness among all the community members. Community partners and the community at large are interested in case management and evidence-based workshops provided by partners through the project. Lessons learned from project will be shared.

**THE EFFECT OF AGING AND NEUROLOGICAL DISORDERS ON MUSCLE FIBER SIZE IN THE PSOAS MUSCLE: A PILOT STUDY.** Arce R, Serrano GE, Russell A, Vargas D, Intorcica A, Walker J, Glass M, Oliver J, Papa J, Nelson C, Sue LI, Beach TG. Banner Sun Health Research Institute; Arizona Alzheimer's Consortium.

Background: Loss of muscle mass, termed sarcopenia, is common in aging and in neurological disorders of aging such as Alzheimer's and Parkinson's diseases, and may affect mobility and performance of daily tasks. It is unknown how these neurological disorders affect the skeletal muscle fiber composition in comparison with normal aging, and how muscles adapt to movement impairment caused by neurological diseases. The purpose of this study was to analyze type 1 and type 2 fibers in the psoas muscle of aging individuals with and without neurodegenerative disorders to gain insight into age and disease-related sarcopenia.

Methods: Psoas muscle was collected at autopsy from human subjects that were part of the Arizona Study of Aging and Neurodegenerative Disorders (AZSAND) and Brain and Body Donation Program (BBDP). Subject selection was based on clinicopathological diagnoses of Parkinson's disease, Alzheimer's disease, progressive supranuclear palsy, and Controls. A longitudinal and cross-sectional portion of the psoas muscle was dissected at autopsy at the level of the L5 of the spinal column and fixed in neutral buffered formalin for two days, then changed to 50% ethanol. Samples were then paraffin-embedded, cut at 6 microns, and mounted on histological slides. Anti-fast and anti-slow myosin heavy chain unconjugated antibodies diluted at 1:3000 with a 97°C sodium citrate antigen retrieval step were used to identify Type I and Type II muscle fiber types. Each section was photographed in three representative areas following staining and a Zeiss AxioVision software was used to determine mean fiber cross-sectional areas.

Results: The group age means were not significantly different, with all groups having a mean of 82 years. Muscle fiber size means for untyped fibers had considerable variation; AD muscle fiber means were smaller than those of controls while PD and PSP means were larger. Type I fibers were similar in the diagnostic groups while Type II fibers were significantly smaller in AD. For PD and PSP, Type II fiber size means were larger than those of controls.

Conclusions: This pilot study showed that Type II muscle fibers may be atrophied in AD but hypertrophied in PD and PSP. Tissue atrophy may explain the findings for AD. For PD and PSP, we hypothesize that tremor and other involuntary muscle movements may induce hypertrophy. Expanding this study to larger subject numbers may provide insight as to how neurological disorders affect skeletal muscles.

**ALZHEIMER'S DISEASE NEUROPATHOLOGICAL COMORBIDITIES ARE COMMON IN THE YOUNGER-OLD.** Beach TG, Malek-Ahmadi M. Banner Sun Health Research Institute; Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: Clinicopathological studies have demonstrated that Alzheimer's disease dementia (ADD) is often accompanied by clinically undetectable comorbid neurodegenerative and cerebrovascular disease that alter the presence and rate of cognitive decline in aging and ADD. Aside from causing increased variability in clinical response, it is possible that the major ADD comorbidities may not respond to ADD-specific molecular therapeutics. As most reports have focused on comorbidity in the oldest-old, its extent in younger age groups that are more likely to be involved in clinical trials is largely unknown.

Methods: We conducted a survey of neuropathological comorbidities in sporadic ADD using data from the US National Alzheimer's Coordinating Center. Subject data was restricted to those with dementia and meeting National Institute on Aging-Alzheimer's Association (NIA-AA) intermediate or high AD Neuropathological Change (ADNC) levels, excluding those with known autosomal dominant AD-related mutations. Subjects were divided into age-at-death categories for analysis: under 60, 60-69, 70-79, 80-89, 90-99 and 100 or over.

Results: Confirmatory of earlier reports, ADD histopathology is less severe with advancing age, effectively increasing the relative contribution of comorbidities, most of which rise in prevalence with age. Highly prevalent ADD comorbidities are not restricted to the oldest-old but are common even in early-onset ADD. The percentage of cases with ADD as the sole major neuropathological diagnosis is highest in the under-60 group, where "pure" ADD cases are still in the minority at 44%. After this AD as a sole major pathology in ADD declines to roughly 20% in the 70s and beyond. Comorbidity rates for some pathologies, especially LBD, are high even in subjects in their 60s and 70s, at nearly 60%, but for most others, their prevalence increases with age. TDP-43 pathology affects more than 35% of ADD subjects 80 and over while microscopic infarcts reach this rate a decade later. Gross infarcts rise more slowly and affect fewer subjects but still involve 15-20% of ADD after age 80. White matter rarefaction may be underestimated in the NACC database but is present in almost 70% of centenarians with ADD.

Conclusions: Effective clinical trials depend on accurate estimates of required subject numbers, which are dependent on observed effect size and clinical response variability. Comorbidities are likely to affect both, leading to lower probability of clinical trial success. Stratifying ADD clinical trial analyses by presence and types of accompanying comorbidities might identify subgroups with higher effect sizes and greater clinical response rates, but accurate in-vivo diagnostic methods for most comorbidities are still lacking.

**CLINICOPATHOLOGICAL CORRELATES OF AGING-RELATED TAU ASTROGLIOPATHY (ARTAG).** Beach TG, Sue LI, Serrano GE, Intorcica A, Walker J, Glass M, Arce R, Oliver J, Nelson C, Russell A, Vargas D, Papa J, Driver-Dunckley E, Mehta SH, Shprecher D, Shill HA, Adler CH. Banner Sun Health Research Institute; Mayo Clinic; Barrow Neurological Institute; Arizona Alzheimer's Consortium.

Background: Aging-related tau astrogliopathy (ARTAG) is a morphologically defined astroglial pathology of elderly individuals with or without neurodegenerative disease. We wished to determine its association with age-related conditions.

Methods: A database search tabulated cases with and without ARTAG diagnosed since 2011 in AZSAND, a longitudinal clinicopathological study of aging in the state of Arizona. Chi-square tests or Fisher Exact tests were used to test for proportional differences while unpaired, 2-tailed t-tests were used to test for differences in means.

Results: ARTAG was present in 189 cases out of 444 specifically examined for its presence. ARTAG was significantly more common in males than females (66%;  $p < 0.01$ ) and the mean age of subjects with ARTAG was significantly greater than that of subjects without ARTAG (88.3 vs 81.6;  $p < 0.01$ ). ARTAG was significantly more common in subjects with mild cognitive impairment (22% vs 12%;  $p < 0.01$ ), vascular dementia (17.5% vs 8.5%;  $p < 0.01$ ), progressive supranuclear palsy (13% vs 5%;  $p < 0.01$ ), argyrophilic grains (48% vs 13%) and a history of traumatic head injury (45.3% vs 31.3%;  $p = 0.016$ ).

Conclusions: ARTAG cases are significantly older than non-ARTAG cases and are significantly more likely to be male than female. ARTAG cases are more likely than non-ARTAG cases to have clinicopathological diagnoses of mild cognitive impairment, vascular dementia, progressive supranuclear palsy and argyrophilic grains. ARTAG cases are more likely to have had a history of at least one traumatic head injury, which increases its previously-noted similarities with chronic traumatic encephalopathy.

**NEUROPATHOLOGICAL DIAGNOSES OF SUBJECTS AUTOPSIED IN THE PHASE 3 CLINICOPATHOLOGICAL STUDY OF FLORTAUCIPIR F18 PET IMAGING.** Beach TG, Montine TJ, Serrano GE, Sue LI, Intorcchia AJ, Walker J, Glass M, Fleisher AS, Pontecorvo MJ, Devous Sr. MD, Lu M, Mintun MA, on behalf of the A16 study investigators. Banner Sun Health Research Institute; Stanford University School of Medicine; Avid Radiopharmaceuticals; Arizona Alzheimer's Consortium.

Background: Avid Radiopharmaceuticals conducted a prospective case-control clinicopathological study of flortaucipir F18 PET Imaging (AV-1451-A16) from October 2015 through June 2018. Sixty-seven valid study autopsies were performed. The cerebral patterns of flortaucipir PET images were visually assessed and compared to the patterns of immunohistochemical tau pathology. The study met pre-specified success criteria, with imaging predicting an NIA-AA B3 level of tau pathology (Braak V/VI) and a high level of Alzheimer's disease neuropathologic change.

Methods: This presentation is an initial report of the detailed neuropathological diagnoses of the 67 primary study subjects. There were 35 females and 32 males, mean age 82.6 (SD 9.4). Fifty-three cases met intermediate or high ADNC levels, consistent with AD as a cause of cognitive impairment. Standardized neuropathological examinations were performed on all subjects.

Results: Many subjects had additional major neuropathological findings (not mutually exclusive), meeting neuropathological diagnostic criteria of dementia with Lewy bodies (DLB; n=7), Parkinson's disease (PD; n=1), progressive supranuclear palsy (PSP; n=5), hippocampal sclerosis (HS; n=5), vascular dementia (n=3) and corticobasal degeneration (CBD; n=1), while others had lesser findings of TDP-43 proteinopathy restricted to the mesial temporal lobe (n=19), Lewy body pathology not meeting criteria for DLB or PD (n=18), age-related tau astrogliaopathy (ARTAG; n=15) or remote cerebral infarcts (n=10). Cases with less than intermediate ADNC (n=14) met neuropathological diagnostic criteria (not mutually exclusive) for PD (n=2), HS (n=2), DLB (n=1), PSP (n=1), CBD (n=1) and others had additional neuropathological findings of TDP-43 proteinopathy restricted to the mesial temporal lobe (n=2), Lewy body pathology not meeting criteria for DLB or PD (n=4), ARTAG (n=3) or remote cerebral infarcts (n=2).

Conclusions: This high proportion of mixed neuropathology is typical of what has been published for other elderly autopsied subjects. Correlations of flortaucipir F18 PET imaging with these varied neuropathological types will be undertaken.

**PROGRESS IN HUMAN FIBROBLAST BANKING PROGRAM AT HUMAN CELLS CORE FOR TRANSLATIONAL RESEARCH AT BANNER SUN HEALTH RESEARCH INSTITUTE.** Beh S-T, Brookhouser N, Brafman D, Serrano GE, Beach TG, Lue L-F. Banner Sun Health Research Institute; Arizona State University; Arizona Alzheimer's Consortium.

Background: The Brain and Body Donation Program (BBDP) at the Banner Sun Health Research Institute (BSHRI) annually receives tissues from 60-90 autopsy cases who were non-demented elderly or had neurological disorders. The Human Cells Core for Translational Research (HCCTR), established since 2018, takes advantage of the BBDP tissue resource to build a human fibroblasts banking program using postmortem scalp tissues. Fibroblasts are widely used for inducible pluripotent stem cells reprogramming and differentiation. The purpose of banking postmortem fibroblasts from clinically and neuropathologically characterized patients is to provide human cells to academic and pharmaceutical communities to facilitate translational research and drug development for age-related diseases that are currently without cure.

Methods: Postmortem human scalp tissues from BBDP donors have been routinely used to obtain fibroblasts by direct culturing of scalp explants. Briefly, scalp tissues (about 1.5-3 grams) obtained at autopsy were immersed in cryoprotectant immediately and stored in refrigerator until processing within 1 week after collection. Dermal tissues were rinsed, chopped into 1-mm<sup>3</sup> pieces, and placed in 6-well plates for explant cultures in a 37°C incubator with 5% CO<sub>2</sub>. Cells were harvested when they reached 80-90% confluence and expanded in T75 culture flasks at 1:2 ratios. Confluent passage-3 cells were harvested, counted, resuspended in cryoprotectant. The cryogenic vials were stored in vapor phase inside a liquid nitrogen tank. The viability of the cells from cryopreservation was evaluated by hemocytometer counting of trypan blue dye-excluded cells. Cell-type-specific protein and gene expression profiling were examined by Western blot analysis, immunofluorescence staining, and qPCR in the passage-3 fibroblasts. Commercially available primary fibroblasts and keratinocytes served as positive and negative cell-type controls, respectively. The apolipoprotein E genotype was determined by qPCR techniques.

Results: Currently, we have banked cryoprotected fibroblasts from 25 cases of donors who were non-demented elderly (N=9) or with clinical diagnoses of mild cognitive impairment (N=3), Alzheimer's disease (N=5), Parkinson's disease (N=5), Parkinson's disease with dementia (N=2), and amyotrophic lateral sclerosis with C9ORF72 gene mutation (N=1). We have observed a consistent pattern during cell outgrowth and expansion. The postmortem fibroblasts maintained high cell viability (90-95%) during cryo-storage. They were characterized by the expression of vimentin, fibronectin, fibroblast-specific protein 1, fibroblast activation protein, alpha-smooth muscle actin, CD73, CD90, CD105, and platelet-derived growth factor receptor beta and absence of epithelial cell marker cytokeratin.

Conclusions: Our results have demonstrated the feasibility of routine banking of patient scalp-derived fibroblasts. The cells exhibited protein and gene expression profiles similar to commercially available primary fibroblasts and maintained high viability in cryoprotectant. Long-term efforts in this cell banking program will result in a valuable human cell resource to use for better understanding of normal aging and age-related neurodegenerative diseases. Our cells have been used for induced pluripotent stem cells (iPSCs) reprogramming by the Arizona State University collaborators. Future use of these cells in direct conversion technology may generate neurons or other brain cell types that could offer the advantage of preserving gene expression profiles and epigenetic properties of the tissue donors.

**CHARACTERIZATION OF MICROSTRUCTURAL, VASCULAR, AND METABOLIC BIOMARKERS IN A PRECLINICAL MODEL OF ALZHEIMER'S DISEASE.** Bergamino M, Scarpelli M, Healey D, Fuentes A, Turner G, Oddo S, Stokes AM. Barrow Neurological Institute; Arizona State University; Arizona Alzheimer's Consortium.

Background: Preclinical mouse models of AD have been developed to recapitulate clinically observed AD features, including amyloid and tau pathology, along with cognitive changes. However, vascular assessment in these models has not been performed, even though it has been increasingly recognized that cerebral vascular changes contribute to Alzheimer's disease (AD) pathology and that these changes occur early in the pathological cascade. Accordingly, the goal of this project is to assess cerebral vascular changes in mouse models of AD that recapitulate human pathophysiology using neuroimaging. We integrate these cerebral vascular changes with assessments of brain microstructure and metabolism. This multi-parametric approach has the potential to shed new insights into AD progression, leading to a broader understanding of the interplay between structural, vascular, and metabolic dysfunction in AD.

Methods: Two groups of mice (wild type (n = 13) and 3xTg-AD (n = 15)) were imaged using multiparametric MRI (Bruker Biospec, 7T) and PET (Bruker Albira) scanners. High-resolution MR images were acquired to quantify volumetric changes in the brain, including the whole brain, hippocampus, cortex, total ventricles, and caudate putamen. Total and microvascular cerebral blood volume (CBV) was quantified using an advanced perfusion imaging method based on spin- and gradient-echo contrast. Diffusion tensor imaging (DTI) data were acquired to quantify microstructural white matter changes. After MRI, mice were injected with <sup>18</sup>F-fluorodeoxyglucose (FDG), and PET images were acquired from 40 to 70 minutes after injection to assess glucose metabolism. On the following day, all mice were sacrificed and brains extracted for genetic and histological analysis.

Results: Preliminary data analysis showed excellent image quality, and advanced metrics within expected physiological limits. A multi-atlas label fusion method has been developed to assess volumetric differences between the groups. Using ROI-based analysis, CBV will be characterized for both total and microvascular perfusion. DTI will be analyzed using DSI Studio, with metrics including fractional anisotropy and axial and radial diffusivities. Glucose metabolism will be compared using ROI analysis of standardized uptake value ratio (SUVR) maps. Data collection and analysis is ongoing, and results will be presented at the meeting.

Conclusions: This study lays the framework for the development of advanced perfusion MRI biomarkers to characterize the neurovascular changes that occur in AD. The cerebral vascular phenotype in these mouse models of AD will be related to structural, microstructural, and metabolic changes using multiparametric MRI and PET. This work represents the first in vivo imaging characterization of these mouse models of AD.

**FREE-WATER DIFFUSION TENSOR IMAGING (DTI) IMPROVES THE ACCURACY AND SENSITIVITY OF WHITE MATTER ANALYSIS IN ALZHEIMER'S DISEASE.** Bergamino M, Walsh RR, Stokes AM. Barrow Neurological Institute; Arizona Alzheimer's Consortium.

Background: White matter (WM) integrity in Alzheimer's disease (AD) can be assessed using diffusion tensor imaging (DTI) with metrics such as fractional anisotropy (FA), axial/radial diffusivities (AxD and RD), and mode of anisotropy. Although standard DTI is susceptible to the effects of extracellular free water (FW), which reduce the accuracy of derived metrics, these effects can be removed using an advanced FW-DTI model. In this study, we investigated differences in WM integrity between AD and healthy controls (HC) using standard and FW-DTI. We hypothesize that FW-DTI will improve the sensitivity and specificity for detection of WM tract abnormalities in AD.

Methods: All data were downloaded from the OASIS-3 brain project database (<http://oasis-brains.org/>). We included 30 HCs (17 females; age (standard deviation) = 73 (6) years; Mini-Mental State Exam (MMSE) = 29.1 (1.2)) and 28 AD subjects (primarily mild AD; 15 females; age = 75 (7) years; MMSE = 24.2 (5.0)). DTI data were preprocessed using FSL. All DTI and FW-DTI metrics were calculated using an in-house MATLAB script. WM integrity was compared between groups using an ANCOVA model, with age and gender as covariates.

Results: No significant differences in age ( $p=0.14$ ) or gender ( $p=0.82$ ) were observed between HC and AD, while significant differences were found for MMSE ( $p<0.0001$ ). Consistent with neurodegenerative mechanisms, lower FA was observed in AD compared with HC mainly in the fornix and corpus callosum (CC) using both standard and FW-DTI. Clusters with seemingly paradoxical increased FA values in AD were found in the anterior thalamic radiation (ATR), cortical spinal tract (CST), and posterior limb of internal capsule (PLIC). These clusters corresponded to regions with more linear anisotropy, suggesting a loss of crossing fiber populations. Widespread increases in AxD and RD were found in AD using standard DTI, while FW-correction yielded significant clusters with both higher and lower AxD and RD in multiple regions with AD pathology. More specifically, FW-AxD was reduced with AD pathology in CC and fornix and predominantly increased in the ATR, CST, and retrolenticular part of internal capsule. Reduced FW-RD was observed with AD mainly in the ATR, CST, PLIC, and superior fronto-occipital fasciculus (SFOF). The FW index may be a useful standalone biomarker of AD-related pathology, with increases in FW indicative of sub-voxel neurodegeneration. Several clusters with higher FW values in AD compared with HC were found, located mainly in the fornix, cingulum, and CC.

Conclusions: The implementation of a FW correction algorithm for DTI can improve the sensitivity and specificity of derived DTI metrics by removing partial volume effects and may better capture underlying AD-related pathologic changes than standard DTI approaches. FW-DTI metrics were more consistent with known AD pathology, both in terms of magnitude and direction of DTI changes. In addition, the FW index may improve sensitivity to sub-voxel neurodegeneration.

**PRECLINICAL EVALUATION OF GLYCOSYLATED PACAP HORMONES IN MICE AND RATS TO TREAT PARKINSON'S DISEASE AND STROKE.** Bernard K, Bartlett MJ, Liu C, Apostol CR, Molnar G, Szabò L, Ronaldson PT, Madhavan L, Streicher J, Heien ML, Polt R, Falk T. University of Arizona; Arizona Alzheimer's Consortium.

Background: The pituitary adenylate cyclase activating polypeptide (PACAP) is an endogenous neuropeptide. PACAP binds and agonizes PAC1, VPAC1, and VPAC2 receptors and inhibits neuronal apoptosis and modulates inflammation. It was shown to be protective in various preclinical models of neurological disorders, including after injection into the striatum in a model of Parkinson's disease (PD) and in stroke models, making it a drug candidate. However, native PACAP exhibits poor pharmacokinetics as it is rapidly degraded; showing low bioavailability.

Methods: We have designed and synthesized several PACAP glycopeptides with various serine glycosides at the C-terminus as well as additional amino acid substitutions to enhance stability. These glycopeptides were evaluated for their ability to stimulate cAMP production in vitro using individual CHO cell lines expressing PAC1, VPAC1, and VPAC2. LC-MS3 was used to test for in vitro stability and blood-brain barrier (BBB) penetration using microdialysis. In order to test for in vivo activity of the lead compound, we started an experiment using a mild progressive unilateral 6-hydroxydopamine (6-OHDA) rat in vivo PD model (n=16/group) tested until 4 weeks post lesion. In addition, transient medial cerebral artery occlusion (tMCAO) studies were done in mice.

Results: PACAP1-27 glycosylation (lactoside among others) maintained receptor agonism with little selectivity between the PAC1 and VPAC1/2 isoforms. Glycosylated peptides showed better stability than native PACAP. In vitro half-life time in CSF matrix glycosylated PACAP ranged from 8.3 min to 11.7 min. In vitro half-life time in water of native PACAP is 2.8 min, which is shorter than all glycosylated derivatives in CSF matrix. In vivo CSF data using 'shotgun microdialysis' coupled with LC-MS3 (n=2, 15 mg/kg, i.p.) shows the PACAP1-27S-Lactoside 2ls98lac is able to penetrate BBB. In the treatment group rats were injected (i.p.) with 15 mg/kg 2ls98lac both 6-hrs prior and 48-hrs post 6-OHDA injection. The cumulative amphetamine-induced rotations at 2 and 4 weeks post-lesion are reduced compared to the vehicle control group (2-tail t-test, p<0.05). Unbiased stereology of dopaminergic (DA) neurons in the substantia nigra (n=8/group) showed in the control group a significant 25% decrease on the lesioned vs the intact side (2-tail t-test, p<0.05), while there was no significant difference between intact vs lesioned side in the PACAP-treated animals. In addition, the tMCAO studies done in mice show that administration (10 mg/kg, i.p.) of PACAP glycopeptides reduced the reperfusion injuries.

Conclusions: Overall, we have observed neuroprotection in vivo and penetration of the BBB, making some of these analogues potential lead compounds for the treatment of neurodegeneration.

Acknowledgement: This work was supported by NINDS RO1NS091238, and the content is solely the responsibility of the authors and does not necessarily represent the official views of the NIH.

**THE EFFECTS OF VOLUME AND LOCATION OF SUBCLINICAL EMBOLIZATION ON THEIR COGNITIVE EFFECTS.** Black J, Murphy D, Wheeler G, Bernstein A, Trouard T, Zhou W. University of Arizona; Arizona Alzheimer's Consortium.

**Background:** Subclinical embolization is common during carotid interventions and percutaneous aortic interventions. Many studies have demonstrated that procedure-related microembolization is associated with post-intervention cognitive decline, however, there is still a significant controversy on the cognitive effects of embolization-related microinfarcts. To understand microinfarcts, we believe that it is important to examine the characteristics of microinfarcts. This study is to examine the relationship between the location and size of subclinical embolization and the long and short-term cognitive changes.

**Methods:** Patients at two academic medical centers who underwent carotid interventions were consented and enrolled. Diffusion-weighted MRIs (DWI) were performed before and within 24-48 hours after intervention to identify procedure-related infarcts. A board-certified radiologist identified lesions as hyperintensities on the diffusion-weighted images that were correspondingly hypointense on the derived apparent diffusion coefficient (ADC) maps. After identification, a semi-automated region-growing algorithm was used to define size and location of microinfarcts using in-house software. Cognitive evaluations were performed pre-intervention, 1 month, and 6 months following intervention to determine short- and long-term cognitive changes. Cognitive scores within each measure were normalized against age and published standard to generate scaled scores. The primary memory outcome measure, Rey Auditory Verbal Learning Test (RAVLT), was normalized against the Mayo Older Americans Normative Studies to generate a MOANS score. Principal component analysis was used to generate a single score for each cognitive measure and spearman correlation was used to determine the influence of location and volume on cognitive function.

**Results:** A total of 181 subjects were recruited and 157 of whom with pre and post DWI images were analyzed. Among them, 77 subjects (49%) had procedure-related microinfarcts with an average volume of 762.90mm<sup>3</sup> (ranging, 18 to 6892mm<sup>3</sup>). Volumes of infarcts were significantly correlated to long-term changes in memory measured by MOANS ( $P < 0.05$ ) and executive function measured Trail Making Test ( $P < 0.05$ ). The most common locations of microinfarcts were parietal and frontal lobes. When the locations of microinfarcts were included in analysis, significant correlations between microinfarcts and changes in cognition were observed in multiple cognitive domains.

**Conclusions:** This prospective longitudinal study demonstrated that both location and size of embolization-related microinfarcts play significant roles in their cognitive effects, suggesting that including lesion characteristics is imperative in evaluating and understanding procedure-related microembolization. Continuing studies will be undertaken to understand how these microinfarcts are associated with CBF changes and disruptions within the brain network topology.

**CELL DEATH AND SURVIVAL PATHWAYS IN ALZHEIMER'S DISEASE: AN INTEGRATIVE HYPOTHESIS TESTING APPROACH UTILIZING -OMIC DATASETS.** Brokaw DL, Piras IS, Mastroeni D, Weisenberger DJ, Nolz J, Delvaux E, Serrano GE, Beach TG, Huentelman MJ, Coleman PD. Arizona State University; Translational Genomics Research Institute; University of Southern California; Banner Sun Health Research Institute; Arizona Alzheimer's Consortium.

Background: Cell death in Alzheimer's disease (AD) is the product of a complex and intercepting network of dynamic molecular pathways. Although multiple programmed cell death (PCD) pathways have been implicated in the neuronal loss observed in Alzheimer's disease, there has been no comprehensive evaluation of the dominant pathway responsible for cell death in AD. Likewise, little research has been done on the relative dominance of survival and PCD pathways in AD or on the potential for cross-pathway interactions.

Methods: Here, we present the results of a hypothesis-driven bioinformatic analysis of PCD and survival pathway activation in paired methylation and expression data from the middle temporal gyrus as well as expression from laser-captured cells from the middle temporal gyrus and hippocampus of AD and control cases.

Results: The results are indicative of PCD pathway activation in AD—of which apoptosis is responsible for the largest fraction of upregulated genes—as well as activation of cell survival pathways.

Conclusions: Collectively, the results hint at a dynamic balance between cell death and survival in AD.

**COMPOSITE SCORE METHODOLOGY COMPARISON FOR A PRE-CLINICAL ALZHEIMER'S DISEASE COGNITIVE OUTCOME.** Brown C, Malek-Ahmadi MH. Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: The Alzheimer's Prevention Initiative Cognitive Composite (APCC) was developed in order to identify a set of sensitive cognitive tests that would be able to detect treatment effects in secondary prevention trials of preclinical Alzheimer's disease (AD). Other cognitive composite scores have used a composite z-score method where the individual tests are transformed into z-scores and the mean of the z-scores is used as the composite score. The APCC method differs in that subtest scores are standardized as a percent of the maximum possible score. While these methods have differing statistical advantages, it is unclear if the two methodologies are comparable in terms of sensitivity to change.

Methods: The first aim of this study was to determine if the APCC composite score method is equivalent to the composite z-score method in terms of its sensitivity to change. The second aim was to determine whether the two methodologies differed in terms of their estimated sample size for a hypothetical AD prevention trial with 25% treatment effect. Data from 414 cognitively unimpaired individuals in the Rush Religious Order Study were analyzed. All individuals carried at least one copy of the APOE ε4 allele. The APCC is comprised of seven cognitive tests that measure episodic memory, processing speed, visuospatial function, executive function, orientation to time, and orientation to place and is calculated by taking the observed score and dividing it by the maximum possible score for each test and then averaging the standardized scores. Using the same set of cognitive tests, we also applied z-score transformations where the raw scores were standardized to the mean and standard deviation of the sample's baseline values for each test. Linear mixed effects models that adjusted for baseline age, sex, education, and baseline composite score were used to estimate the mean and standard deviation of annualized change. The mean to standard deviation ratio (MSDR) was calculated for both methods and compared where higher MSDR values indicate greater sensitivity to change.

Results: The APCC composite methodology yielded a mean annualized change of  $-1.25 \pm 1.65$ , 95% CI: (-1.41, -1.09). For the z-score composite methodology, the mean annualized change was  $-0.11 \pm 0.15$ , 95% CI: (-0.13, -0.10). The MSDR for the APCC composite methodology was 0.76 and 0.75 for the z-score composite method respectively. The sample size estimate for a 25% treatment effect of the MSDR for the APCC methodology  $n = 424$  per treatment arm while the z-score method indicated a sample size  $n = 427$  per treatment arm.

Conclusions: These findings indicate that the APCC and z-score methods for calculating cognitive composite scores are comparable in terms of their sensitivity to change and estimated sample size for secondary prevention trials. Future studies are needed to determine the robustness of each method with regard to outliers and missing data. Additional analyses using mixed model repeated measures (MMRM) analyses will help determine how these methodologies may perform in statistical method commonly used in clinical trials.

**TRAINING AND DISSEMINATION OF MODEL-INFORMED DRUG DEVELOPMENT AND REGULATORY COMPLIANT DATA MANAGEMENT IN ALZHEIMER'S RESEARCH FOR THE AAC COMMUNITY.** Burton JK, Hanan N, Sinha V, Sivakumaran S, Romero K on behalf of the Critical Path for Alzheimer's Disease. Critical Path Institute; Merck & Co. Inc.; Arizona Alzheimer's Consortium.

Background: The Critical Path for Alzheimer's Disease (CPAD) consortium continuously updates and enhances regulatory-endorsed quantitative drug development tools (QDDTs). These tools are hosted in clinical trial simulation platforms, based on comprehensive drug-disease-trial models, which provide quantitative descriptions of relevant aspects of disease progression, drug effects, placebo effect and dropouts. The quantitative tools allow researchers to optimize the design of clinical studies, by making more informed decisions regarding sample size estimation, trial duration, frequency of observations, total time of follow-up, specific design approaches, entry criteria, enrichment strategies and stratification approaches. The goal of this work is to develop training platforms that will provide didactic content to AAC researchers on the use of quantitative drug development tools developed by CPAD, and regulatory grade data standards for non-clinical and clinical studies. The envisioned learning objectives for audiences will be to gain an understanding of the concepts and use of regulatory-endorsed clinical trial simulation tools in AD to address relevant challenges in AD drug development.

Methods: The content for the QDDT training platform will utilize different media to provide effective dissemination of material to wide audiences. Such media include traditional slide-based presentations, live hands-on demos of the tools, animated videos, and written documentation. The content in this platform is intended to provide an overview of the conceptual methods behind the simulators, followed by an overview of the simulation environment of the GUI, and how to interpret the simulation outputs. All material is intended for wide audiences with no specific training in quantitative sciences. The proposed training course on the adoption and implementation of regulatory-grade data standards for non-clinical and clinical research will utilize a similar multimedia approach. The content in this platform is aimed at researchers of all backgrounds, regardless of their knowledge of data standards, such as CDISC, SDTM, or ADAM. The training will be focused on the annotation of CRFs and laboratory notebooks, as well as basic concepts of relational table data structures, and the generation of analysis subsets.

Results: A communication framework was developed that outlines the key concepts and messaging for both platforms. This framework is being used to generate didactic content that includes presentations, videos, and written documentation. For the QDDT training platform, various case studies focused on specific and realistic study design challenges for drug development and observational studies are being used to highlight how previously developed tools can be leveraged. The platform will address questions regarding study design, which include the definition of entry criteria, enrichment strategies, stratification approaches, sample size estimations, balancing of power and attrition, definition of hypotheses for statistical inference and their impact on analysis approaches, and impact of study power based on trial duration. In parallel, the underlying data used to generate the QDDTs will serve as case studies for the training platform on regulatory grade data standards.

Conclusions: The envisioned outcome of the training platforms being developed will allow wide audiences to gain a deep understanding of the concepts, advantages, and limitations of data-driven QDDTs. The specific case studies leverage existing QDDTs for Alzheimer's disease in terms of clinical trial design optimization. The totality of work represents an end-to-end approach for addressing the challenges in AD drug development, thereby helping to accelerate the development of novel therapies.

**APOE EFFECTS ON RESTING STATE NEURAL NETWORKS IN COGNITIVELY NORMAL ADULTS.** Cabello-Arreola A, Baxter LC, Zhou Y, Hu L, Caselli RJ. Mayo Clinic Arizona; Arizona Alzheimer's Consortium.

Background: Memory decline prior to age 60 was previously observed in a presymptomatic cohort of APOE e4 carriers (Caselli et al., 2009). Standard imaging evaluation cannot readily detect structural changes in APOE e4 individuals. Network integrity may be a more sensitive measure of subtle cognitive changes in otherwise in asymptomatic carriers. Resting state functional connectivity (rsFC) methods allow investigation of the integrity of whole brain neural networks. In this preliminary study, we investigated the effects of the APOE e4 allele on rsFC networks in well-characterized, cognitively intact adults as part of larger study determining the influence of genetic heterozygosity on neural functional and structural integrity and cognitive aging and dementia.

Methods: This preliminary data includes 38 adults aged 40 to 80 years (age mean  $\pm$ SD: e4 carriers=64.9 $\pm$ 11.27, noncarriers=65.4 $\pm$ 7.85; p-value=0.87) who are participating in the longitudinal APOE cohort study. Resting state fMRI imaging was obtained with the following parameters: TE = 27ms, TR = 2000 msec, resliced to slice thickness = resulting in 2x2x2 mm<sup>3</sup> voxels, 120 volumes collected. The fMRI data was analyzed using an independent component analysis (ICA) technique (GIFT), thus identifying different components (i.e. brain networks). We isolated 20 ICA components from the initial ICA analysis. T-tests of group differences based on APOE status (e4 carrier vs. noncarrier) were used to determine relative integrity of network functional connectivity.

Results: Visual inspection of components identified commonly observed brain networks identified through rsFC techniques, including the default mode network (DMN; posterior and anterior divisions), executive control, frontoparietal (FPN), auditory, sensorimotor, vision (inferior and medial divisions), and temporal networks. Correlations between average spatial functional network connectivity (FNC) networks of  $> .50$  were used to indicate strong network relationships and  $< .30$  low correlations. The APOE noncarriers showed greater connectivity of vision-language network while APOE e4 carriers had no network pairs showing the same degree of interconnectivity. In contrast, the e4 carrier group showed lower network interconnectivity for 3 pairs of regions: temporal-medial vision, auditory-medial vision, and FPN-medial vision network pairs compared to the noncarrier group.

Conclusions: In this preliminary analysis, we found less interconnectivity of resting state networks in a group of APOE e4 carriers compared to noncarriers, especially frontal connectivity with other areas. This suggests that network interactions necessary to effectively carry out higher cognitive functions such as memory may be compromised in asymptomatic e4 carriers. rsFC analyses may be sensitive to changes in brain integrity well before detection by other methods and may provide better detection of early regional brain compromise.

**IMPLEMENTATION OF A PERSON CENTERED CARE INTERVENTION: DO YOU KNOW WHO I AM?** Cain A, Detaranto B, Razo S, Smith J, Johnson K. HonorHealth Thompson Peak Medical Center.

**Background:** Hospitalized older adults present with diverse needs and cognitive and functional limitations. Person-centered care (PCC) is an approach to help personalize interventions among older adults with cognitive and functional limitations. PCC includes knowing the individual, his or her needs and preferences to provide empathic care. The 2018 Alzheimer's Association Dementia Care Practice Recommendations include PCC as a philosophy of care developed around the needs of the individual and knowing the person. **Methods:** The purpose of this project was to evaluate whether introduction and implementation of a person centered care (PCC) intervention using an "All About Me Board" (AAMB) tool can 1) change workplace climate perception among registered nurses (RN) s and patient care assistants (PCA) s during patient interactions, 2) decrease falls, and 3) increase patient satisfaction. The AAMB were placed in a patient's room and made visible to the patient and for staff to use with patients. Staff were educated on the purpose and how to use to personalize patient care and interactions using the AAMB education protocol. The nurse caring for the patient explained the purpose of the AAMB to the patient and family. Patients and family members were asked to add to or complete items on the AAMB and if family was not available, nursing were asked to add to or complete items on the AAMB. Staff were asked to present one item from the AAMB during shift report. Approach strategies for healthcare personnel who entered the patients room included 1) always introduce yourself, 2) smile and have a warm demeanor, 3) get to the patients eye level, 4) use a pleasant voice, 5) go slow, talk in short simple sentences, 6) validate and redirect attention, 7) appeal to the emotion, and 8) let the person know you will keep him or her safe. Participants who met inclusion criteria were asked to participate in the pre-test/post-test survey. Education was mandatory for all RNs and PCTs as the AAMB tool was standard of care for patients on the study unit. Participants were given two weeks to complete the pre-test survey and place in the return box located in the study unit. Upon completion of the pre-test survey, the co-investigators provided education about PCC; "Individualizing care among patients; knowing the patient first, rather than the disease can be successful with patient safety" and reviewed use of the of the AAMB tool for all RNs and PCTs employed on the medical surgical unit. To determine support of the AAMB program and understanding of PCC, study participants were asked to complete the same post-test survey three months post intervention which were distributed in the same manner as the pre-test survey. **Results:** There were a total of 26 (88.3%) RNs and 5 (17.2%) PCTs who participated. The majority of participants were female 25 (83.3%) between the age of 25 - 34 years 12 (40%). The majority of RNs held a Bachelor's Degree in Nursing 18(60%) with the majority of participants working from 7 a.m. to 7 p.m. and with one to five years of experience 12(40%). Workplace climate perception were measured pre and post intervention. For the variable "I experience my workplace as a place where it is neat and clean" (F 4.085, df 1.000 p < .048), "I experience my workplace as a place where it is easy for patients to keep in contact with their loved ones" (F 5.990 df 1.000, p < .017), and "I experience my workplace as a place where patients have someone to talk to if they wish" (F 6.682, df 1.000 P < .012). There were no falls and falls with injury three months pre implementation. Three months post implementation there were three falls without injury, with one fall occurring each month (August, September, October 2019) and no falls with injury. The two HCAHPs questions measured three months pre implementation and three months post implementation for the questions, "During this hospital stay how often did nurses listen to you" and "During this hospital stay how often did doctors listen to you"? Three months pre implementation for "nurses listening to you" a range from 84% to 86% for always, and for "physicians listening to you" a range from 78 to 87% were noted. Post implementation range for nurses were from 66% - 95% and physicians 61% -95% for always. **Conclusions:** Knowing your patient involves a patient centered strategy where health care providers can individualize patient care for optimal engagement.

**AGE-RELATED, SPECIFIC CHANGES IN EXPRESSION OF SEVERAL CENTRAL MELANOCORTIN RECEPTOR SUBTYPES IN THE RAT.** Chawla MK, Zempare M, Hruby V, Barns CA, Cai M. University of Arizona; Arizona Alzheimer's Consortium.

Background: The five melanocortin receptors cloned so far (MCR1-5) have been associated with control of inflammatory disorders, immunomodulation, antipyretic effect and prevention of brainstem ischemia and reperfusion injury (Schimolli et al. 2009). It has been reported that the human melanocortin 4 receptor (hMC4R) is involved in neurodegenerative disease (Shen et al., 2016). Melanotropins may protect against the progression of Alzheimer's disease (Giuliani et al., 2014). Furthermore, administration of  $\alpha$ -MSH or its more stable analog [Nle<sup>4</sup>,D-Phe<sup>7</sup>]- $\alpha$ -MSH (NDP- $\alpha$ -MSH) has been observed to enhance learning and memory (Beckwith, et al. 1975). However, the impact of age with respect to melanocortin receptor expression remains unexplored. Previously we have shown that the total expression of melanocortin receptor is reduced in the aged rats in four of the six brain regions studied.

Methods: In the present study, we systematically investigated the expression of 5 different subtypes of melanocortin receptors in brain of young (9 months) and aged (23 months) rats that were assessed for their cognitive status in memory tasks. Spatial memory and visual discrimination ability were assessed using the Morris watermaze task. Six regions of the brain were extracted from each animal, including the frontal cortex + anterior midbrain, parietal cortex, cerebellum, posterior midbrain, hippocampus and occipital lobe. We collected the membrane fragments from each region of all animals in each age group, then ran a specific binding assay using iodine labelled NDP- $\alpha$ -hMCHR1-5 on a high throughput Micro Beta II radiation counter. Six samples were measured from each animal for each region, and then averaged to produce a single count for each animal in each region. All measurements were collected in a blind fashion. We then ran a linear regression analysis with spatial learning behavior and specific receptor binding for hMCHR1-5 receptors using Graph-pad Prizm software.

Results: A significant correlation was found between spatial memory and two receptor subtypes MCH-1R ( $p = 0.048$ ) and MCH-3R ( $p = 0.049$ ) in old animals.

Conclusions: This finding potentially opens a new window of discovery for exploring and developing new treatments for cognitive changes that arise in normative aging and in neurodegenerative disease.

**ASSOCIATION OF PULSE BLOOD PRESSURE WITH FLORTAUCIPIR-PET MEASURED TAU DEPOSITS IN ENTORHINAL CORTEX IN COGNITIVELY UNIMPAIRED INDIVIDUALS.** Chen K, Malek-Ahmadi M, Protas H, Ghisays V, Chen Y, Devadas V, Goradia D, Lee W, Luo J, Caselli RJ, Su Y, Reiman EM. Banner Alzheimer's Institute; Mayo Clinic Arizona; Arizona Alzheimer's Consortium.

Background: Various cardiovascular risk factors have been examined for their potential role for Alzheimer's disease (AD). In this study, we examined the relationship between pulse blood pressure (pBP) and tau deposits in entorhinal cortex in cognitively unimpaired (CU) individuals who have 0, 1 or 2 copies of APOE4 allele. pBP is defined as the difference between the systolic and diastolic blood pressures.

Methods: Data from total 122 study participants in Arizona APOE cohort were included in this report. There were 21 APOE4 homozygotes (62±8 years old, 85% female), 36 heterozygotes (68±9, 67%) and 65 non-carriers (67±8, 82%). Tau-deposits measured by flortaucipir PET and quantified as standard uptake value ratio (SUVR) were obtained for all subjects with partial volume correction using the geometric transfer matrix technique and the cerebellum as the reference region. Blood pressures were measured 3 times during the patients had their PET scans: prior to the IV-placement, prior to the tracer injection and at the end of the scan. The three systolic and diastolic measures were separately averaged and used to form pBP. General linear model (GLM) with pBP, APOE4 dose, age and sex as predictors and entorhinal cortex flortaucipir SUVR as dependent variable was used to assess the association of pBP with flortaucipir SUVR, with each of the rest of the covariates and their interactions with pBP. Post-hoc analysis was also performed to the association of pBP with inferior temporal flortaucipir SUVR using the same GLM.

Results: Except age (62±8, 68±9, and 67±8 for HM, HT and NC,  $p=0.01$ , ANOVA), the 3 APOE4 groups were no different in sex ratio, education, MMSE and memory performances ( $p>0.05$ , ANOVA). pBP was positively associated with entorhinal cortex flortaucipir SUVR ( $p=0.0102$ ) in the presence of significance interaction of pBP with age ( $p=0.0114$ ) and marginally significant interaction of pBP with APOE4 ( $p=0.0757$ ). In the post-hoc analysis, no significant association was found between inferior temporal flortaucipir SUVR and pBP but in the presence of significant interaction of pBP with APOE4 ( $p=0.0035$ ).

Conclusions: Increased pBP is associated with higher tau deposition in the entorhinal cortex accounting for the effects of age, APOE4 dose and sex in CU individuals. Further and larger studies are needed to confirm our preliminary findings.

**FEASIBILITY STUDY: MAPPING MOCA ITEM-LEVEL SUB-SCORES TO MMSE TOTAL SCORE BASED ON MACHINE LEARNING TECHNIQUES.** Chen K, Malek-Ahmadi M, Atri A, Beach T, Saner D, Bauer R, Devadas V, Su Y, Reiman EM. Banner Alzheimer's Institute; Banner Sun Health Research Institute; Arizona Alzheimer's Consortium.

Background: The most-widely used and long-standing Mini-Mental State Examination (MMSE), and the relatively recently introduced and increasingly used Montreal Cognitive Assessment (MoCA) are two of the most commonly-used screening tests for cognitive impairment. To reduce the possible confusion of two similar general cognitive tests for clinicians and to harmonize data across different datasets for joint analysis, we investigated the feasibility of mapping item-level MoCA data to the MMSE using machine learning (ML) techniques.

Methods: Data of 1982 study participants from ADNI, the Longevity Study, and the Brain and Body Donation Program (BBDP) together with 3 different item-level subscore arrangements were included in this study. Based on the previously reported difficulties of further improving the total-score relationship between MoCA and MMSE (Bergeron, et al, JAGS, 2017), we attempted to establish a mapping from the item-level scores of the MoCA to the MMSE total score. Three ML algorithms, Partial least square (PLS), regression tree ensembles and neural network analysis, were each evaluated and compared for their capacity to establish a robust and accurate mapping of MoCA scores to MMSE scores. A commonly adopted cross-validation scheme was used by randomly dividing data into training, validation, and testing sets for each ML approach. Performance was assessed using the correlation coefficient between the estimated and measured MMSE total scores for individuals in the testing dataset.

Results: The simple correlation coefficient between MMSE total and MoCA total is 0.76. When MoCA item-level sub-scores were used to estimate MMSE total score, however, this correlation coefficient increased to 0.87 for PLS-based and 0.89 for regression tree ensemble. For a neural network with 6 neurons and 12 neurons separately for layers 1 and 2, the correlation coefficient was 0.91 between the measured and the MoCA sub-score based MMSE total score estimates.

Conclusions: The mapping established in this preliminary study demonstrated its adequacy and robustness in using item-level MoCA scores to predict MMSE performance. These findings could potentially be improved when larger datasets are available and after the settings for neural network and regression tree ensemble algorithms are optimized more comprehensively. Ultimately, these methods can be used to provide clinicians and researchers with a reliable, data-driven approach to translating between MoCA and MMSE scores.

**VISUALIZATION AND QUANTIFICATION OF SPATIAL PATTERN OF HYPOMETABOLISM OR GRAY MATTER ATROPHY IN INDIVIDUAL PATIENTS: ASSISTING CLINICAL DIAGNOSIS OF ALZHEIMER'S DISEASE.** Chen K, Huang Q, Liu X, Ge X, Lee F, Cheng B, Su R, Wang J, Hall L, Chen W, Reiman EM. Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: Researchers from Banner Alzheimer's Institute previously reported a SPM-based computer package to characterize the spatial pattern of FDG-PET measured glucose hypometabolism of an individual patient in comparison with a group of cognitively unimpaired (CU) normal subjects (Chen, et al., SPIE Medical Imaging, 2006). The consistency of such spatial pattern in a given individual patient with the specific one for AD patients can be informative in assisting physicians to make a clinical diagnosis. Supported by Shanghai Green Valley Pharmaceutical, LLC, we extended this package capacity to process MRI data, and made it operating-system independent.

Methods: We updated AD specific spatial pattern of either hypometabolism for FDG-PET or regional gray matter atrophy for T1-MRI by comparing 190 AD patients with 190 CU subjects from ADNI. Patterns of a given individual patient, or the AD group can be visualized with or without the multiple comparison correction per physician preference. In addition, either hypometabolic or regional gray matter atrophy patterns can be superimposed on top of the patient's high-resolution T1-weighted MRI, if it is available. For FDG-PET, such display provided with complementary structural information in the patient's own brain coordinate space. Furthermore, the degree of pattern consistency between the AD group and a given patient now can be quantified using the hypometabolism convergence index (HCI, Chen, et al., Neuroimage, 2011) for FDG-PET. Finally, bimodal PET/MRI data can be jointly processed, analyzed and displayed.

Results: Based on CAT12 pipeline in SPM12, the package can run on Windows, Mac and Linux platforms. Data from multiple patients can be batched. As an example, the figure shows the hypometabolism pattern in blue color at FWE corrected  $p=0.05$  for one patient together with the AD specific hypometabolism pattern outlined as red boundaries.

Conclusions: The procedure is fully automated. It can be helpful in clinics providing assistances to physicians who see patients routinely for AD diagnosis. We are working to incorporate the convolutional neural-network based classification as additional clinical diagnosis assistance as part of the package. We are also working on the package to enhance its capacity to process, analyze and visualize other PET and MRI data types.

**ALLOPREGNANOLONE PROMOTES NEURAL STEM CELLS DIFFERENTIATION TO NEURONS AND OLIGODENDROCYTE PRECURSOR CELLS.** Chen S, Wang T, Yao J, Brinton RD. University of Arizona; University of Southern California; Arizona Alzheimer's Consortium.

Background: Allopregnanolone (Allo), an endogenous neurogenic steroid, promotes proliferation of human and rodent neural progenitor/neural stem cells (NSCs) in vitro, reverses neurogenic and cognitive deficits in the triple transgenic mouse model of Alzheimer's disease (3xTgAD) in vivo. In this study, we investigated the impact of Allo on neural differentiation in vitro and in vivo of 3xTgAD.

Methods: In this study, we first determined the age- and AD-gene related decline in neuronal differentiation capacity in cultured NSCs and fixed tissue sections derived from age-matched 3xTgAD and nonTg mice brains. Then we investigated the impact of Allo treatment on neural differentiation in cultured NSCs from adult mouse in vitro via immunocytochemistry analysis of cell type specific labeling. We further investigated the efficacy of Allo on promoting neuronal differentiation in 5-month-old male 3xTgAD mice. We conducted these in vivo studies using flow cytometry analysis, immunoblot and immunohistochemistry analysis.

Results: We found an age- and Alzheimer's-dependent decrease in overall NSCs differentiation with a shift from neuronal to glial differentiation. Consistently, the number of immature doublecortin (DCX) positive neurons declined more significantly with aging in 3xTgAD mice compared to age-matched nonTg mice. Allo treatment significantly increased the ratio of MAP2-positive-neurons to GFAP-positive-glias from cultured adult mouse NSCs. To further determine the efficacy of Allo to prevent the loss of neuronal differentiative capacity in vivo, we investigated the impact of Allo on neural differentiation in 5-month-old male 3xTgAD mice. Flow cytometry-based analysis indicated that Allo treatment increased the number of newly generated neurons as indicated by the increase in BrdU/NeuN double positive cells. Immunostaining on brain sections confirmed that the number of DCX positive neurons was significantly increased following Allo treatment, which was further supported by the enhanced level of DCX expression in hippocampal samples. Allo treatment also increased the expression of Olig2, an oligodendrocyte precursor cell marker. Immunohistochemistry analyses showed that more Olig2 positive cells were distributed in corpus callosum area in the Allo-treated brain. The increase of differentiation in neuronal cell and oligodendrocyte precursors was paralleled with an increase of expression in insulin-like growth factor-1 (IGF-1) and IGF-1 receptor (IGF-1R).

Conclusions: These findings suggest that Allopregnanolone is a regenerative therapeutic candidate to prevent or delay neurogenic deficits associated with age and Alzheimer's disease.

**COLLABORATIVE GLOBAL-LOCAL NETWORK FOR CLASSIFICATION OF PATIENTS WITH ALZHEIMER'S DISEASES AND COGNITIVELY UNIMPAIRED INDIVIDUALS: A NEURODEGENERATION ASSESSMENT TOOL ASSISTING FOR CLINICAL DIAGNOSIS AND PATIENT SCREENING IN AD TRIAL.** Chen W, Yuan Y, Reiman EM, Liu X, Lee F, Wang Z, Chen K. Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: MRI (magnetic resonance imaging) based neurodegenerative assessment has been commonly used in assisting the clinical diagnosis for Alzheimer's disease (AD) and in screening patients for inclusion in clinical trials. Deep Learning (DL) and Computer Vision based methods are recently proposed to distinguish patients with AD from cognitively unimpaired (CU) individuals using MRI data. Most methods, however, leverage the whole brain image as the only input, not explicitly exploiting few key brain regions, critical to the disease's development. One such brain region is the hippocampus, an area which has been routinely used in routine clinical diagnosis for its regional atrophy. While some whole brain image features extracted by DL are as critical, between-subject variance for other less relevant brain areas probably contributed less to the AD/CU distinction, caused overfitting, and might serve as 'noise'. We therefore introduce an approach, used in previous visual attention studies, that combines both local and global information.

Methods: We used 1973 T1 MR images from 1079 ADNI individuals, and randomly divided 80/10/10% of the data to training/validation/testing datasets. A collaborative global-local network of multiscale model was designed to use both local fine structures and global features. We especially in this study used left and right hippocampus sub-images as separate inputs, preserving local information in original full resolution. Additionally, the whole brain MRI was down sampled to a lower resolution balanced between reducing the 'noise' effects and keeping useful features which can be unearthed via DL. The three inputs were fed into three independent branches of 3D convolution layers first, and a late-fusion collaboration strategy was utilized to fuse features from them for AD classification.

Results: Based on the results from the testing dataset, we achieved 89.8% classification accuracy (TN rate = 91.0% and TP rate = 88.6%). When using only the global data (i.e., the whole brain MRI only), the classification accuracy was 86.3% (TN = 89.7%, TP = 82.9%)

Conclusions: We provided evidence that collaborative leverage of both local and global information is important for accurate AD classification. Future studies are needed to confirm our findings.

**SELF-PU: SELF BOOSTED AND CALIBRATED POSITIVE-UNLABELED DEEP LEARNING MODEL TRAINING.** Chen X, Chen W, Chen T, Yuan Y, Gong C, Chen K, Wang Z. University of Science and Technology of China; Texas A&M University; Nanjing University of Science and Technology; Banner Alzheimer’s Institute; Arizona Alzheimer’s Consortium.

Background: Many real-world applications of deep learning (DL) have to tackle the Positive-Unlabeled (PU) learning problem. That is, learning a binary classifier from a large amount of unlabeled data and a few labeled positive and negative examples.

Methods: While current state-of-the-art methods employ importance reweighting to design various risk estimators to deal with the existence of the unlabeled data, they do not take advantages of the learning capability of the DL model itself, which could provide reliable supervision. We propose a novel Self-PU learning framework, which seamlessly integrates PU learning and self-training. Self-PU highlights three “self”-oriented building blocks: a self-paced training algorithm that adaptively discovers and augments confident positive/negative examples as the training proceeds; a self-calibrated instance-aware loss; and a self-distillation scheme that introduces teacher-students learning as an effective regularization for PU learning.

Results: In comparison to well-known algorithms, we demonstrate the superior performance of Self-PU using database such as MNIST (<http://yann.lecun.com/exdb/mnist/>) and CIFAR10 (<https://www.cs.toronto.edu/~kriz/cifar.html>),. Moreover, we applied Self-PU to distinguish patients with Alzheimer’s disease and cognitively unimpaired individuals using 3D T1 weighted magnetic resonance imaging data from Alzheimer’s Disease Neuroimaging Initiative (ADNI) and found that Self-PU obtains significantly improved, more accurate results over existing methods.

Conclusions: Self-PU takes advantages of the DL model self-learning capacity to deal with the unlabeled data and is with potential to make the DL application in our AD research more powerful.

**THE EFFECTS OF TMEM184B OVEREXPRESSION ON ALZHEIMER'S DISEASE MEMORY DECLINE.** Cho TS, Hart HR, Wright EB, Bhattacharya MRC. University of Arizona; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease (AD) is a neurodegenerative disease characterized by the progressive impairment of cognitive functions such as memory. One pathological change resulting from AD is axonal dystrophy, which is associated with the extracellular depositions of amyloid  $\beta$  ( $A\beta$ ). Tmem184b is highly expressed in the hippocampus and cortex, and it contributes to axon degeneration and synaptic dysfunction, suggesting its activity could promote AD pathology.

Methods: We intracranially injected custom-designed adeno-associated virus to over-express Tmem184b in AD-model (5xFAD) mouse hippocampi. Novel object recognition assays were performed to assess memory. We also utilized immunohistochemistry.

Results: Here we will present our preliminary findings on the effect of Tmem184b over-expression on short-term and long-term memory using the novel-object recognition assay between two and six months of age. We will also present immunohistochemical analysis evaluating whether Tmem184b overexpression influences astrocyte or microglial activation, or plaque number.

Conclusions: The data we have collected thus far reveals a potential role of Tmem184b in AD-associated decline.

**A NEW APPROACH TO THE CELL/MOLECULAR BIOLOGY OF ALZHEIMER'S DISEASE.**  
Coleman PD, Mastroeni D, Brokaw D, Delvaux E, Huseby C. Arizona State University; Arizona Alzheimer's Consortium.

Background: The multiple failures of drug trials in Alzheimer's disease suggest that a new model of the biology of this disease is needed. We here present data supporting a central role for the nuclear pore complex in neurons in early stage of disease.

Methods: Postmortem human brain tissue was examined using methods of immunohistochemistry, in situ hybridization, RNAseq, biomathematical analyses. Also in vitro experimental methods.

Results: Results show disruption of exchange of molecules between the cell nucleus and cytoplasm of neurons as an early phenomenon in the cellular course of disease. This disruption is attributable to changes in both the nuclear pore complex itself as well as molecules that have roles in facilitating transport between nucleus and cytoplasm. One class of molecules whose transport is affected early in disease are epigenetic molecules that play critical roles in chromatin structure and, thus, availability of genes for transcription that are central to the neuronal phenotype of Alzheimer's disease.

Conclusions: Data presented support a model of changes in the cell/molecular biology of individual neurons that centers around the role of transport between cell nucleus and cytoplasm of neurons as an early phenomenon in Alzheimer's disease that may account for myriad changes in gene expression and the neuronal phenotype known to occur in this disease.

**SUCCESSFUL RECRUITMENT OF EARLY-STAGE ADRD PARTICIPANTS THROUGH COMMUNITY PARTNERSHIPS AND HIPAA-COMPLIANT FAX REFERRAL PROCESS.**  
Cortés M, Angulo A, Glinka A, Gonzalez MS, Stotler K, Coon DW. Arizona State University; Arizona Alzheimer's Consortium.

Background: The EPIC (Early Stage Partners in Care) II study is a randomized clinical trial of a psychoeducational skill-building intervention that targets the relative lack of stage-appropriate community services for dyads where one person (EP) is in the early stages of ADRD and the other (CP) is the current or future care partner. EPIC II translates EPIC (a manualized early-stage, group dyadic intervention that was embedded in the community from its inception) across urban and rural communities in Arizona and Nevada, which have the first and third highest projected rates of AD increase by 2025 – 42.9% and 36.2% respectively. Recruitment and delivery of EPIC is a coordinated effort of an academic research institution with local Alzheimer's Association Chapters, area agencies on aging, a promotores network, other community-based organizations, and the Arizona Alzheimer's Disease Center. Recruitment strategies were modeled on the pilot study's successes, utilizing the extensive network of research, healthcare, and community organizations with which the project team has firm ties. The cornerstone of successful recruitment has been a HIPAA-compliant, IRB-approved fax referral process used among partners, community organizations, and potential participants. This referral process reduces referral lag time by allowing community partners to obtain and document up-front permission from interested individuals who wish to be contacted by the study team for screening. Prior research shows the average length of time for individuals to follow up on a received referral is 28 months, and typically after the person with ADRD has progressed beyond early-stage. Therefore, close partnerships with community providers and early contact with potential participants is crucial to study success.

Methods: This presentation reviews all 256 EPIC II study referrals received from October 2017 through February 2020. The data were analyzed to characterize referral sources and compare the outcome of referrals by source.

Results: Of the total number of referrals (N=256), fax referrals from community partners made up the largest proportion (73.0%), followed by referrals from an Alzheimer's registry (13.7%) and self-referrals (13.3%) from interested participants who contacted the team after learning of the study through online resources, public conferences, or word of mouth. In the first two years, 85.9% of referrals were received through HIPAA-compliant faxes from community partners, while in the third year that proportion dropped to 29.3% and referrals from the registry made up 60.3% of the total referrals. Still, a slightly higher, but non-significant, proportion of referrals from providers working with participants (38.0%) enrolled in the study versus those from participants themselves (35.3%). The proportion of AD registry referrals that enrolled in the study (14.3%) was significantly lower than from the other two methods; however, it is important to note that the Alzheimer's registry was a new recruitment strategy implemented in the third year of the study.

Conclusions: Findings are consistent with the pilot study's successful recruitment using the HIPAA-compliant fax referral process with community partners. In addition, the data highlight that a key community partner engaged directly with clients is necessary in combination with this process. While participants who are self-directed in seeking out resources are highly likely to end up enrolling in research, they are a relatively small proportion of the overall referrals. The registry has shown promise as a new strategy that could serve to fill a critical gap going forward, even though the proportion of referrals that end up qualifying and enrolling is comparatively low.

**SURFACE-BASED MORPHOMETRY ANALYSIS FOR STUDYING VENTRICULAR ABNORMALITIES OF COGNITIVELY UNIMPAIRED SUBJECTS PRIOR TO CLINICALLY SIGNIFICANT COGNITIVE DECLINE.** Dong Q, Zhang W, Stonnington CM, Wu J, Gutman BA, Chen K, Su Y, Baxter LC, Thompson PM, Reiman EM, Caselli RJ, Wang Y. Arizona State University; Mayo Clinic Arizona; Illinois Institute of Technology; Banner Alzheimer's Institute; Barrow Neurological Institute; University of Southern California; Arizona Alzheimer's Consortium.

Background: Ventricular enlargement is associated with AD-related neuropathological progress, and abnormal ventricular volume (VV) expansions have been detected prior to cognitive decline, as measured by neuropsychological tests and carefully conducted interviews. However, current VV measures do not pinpoint the details of subregion expansions. To decode the exactly deformative ventricular subregions in the preclinical stage, this work described a completely automated ventricular morphometry analysis system (VMAS) which generates a whole connected 3D ventricular shape model suitable for the ventricular morphometric analysis.

Methods: VMAS contains a novel automated ventricular segmentation approach and a statistical evaluation of multivariate tensor-based morphometric (mTBM) features. Firstly, individual MRI scans were linearly registered into a standard space (MNI152). Secondly, the registered images were segmented into three brain tissue types (the gray matter, white matter, and CSF); a group-wise CSF template was created from all individual CSF masks, and the group-wise ventricular template was subsequently obtained from the group-wise CSF template with the geodesic shooting algorithm. Thirdly, binary ventricular masks were segmented and extracted, then ventricular surface meshes were constructed and smoothed. Fourthly, the whole ventricular surface was cut into three sub-structures via a holomorphic flow segmentation method. Fifth, each sub-ventricular surface was conformally mapped to a rectangle in the parameter domain, and vertex-wise mTBM statistics were estimated on these mapped surfaces. Finally, the morphometric variations of ventricular surfaces between groups were evaluated, and significantly different subregions were shown in the form of a p-map of the comparison analysis.

Results: We applied VMAS to two independent datasets of normally aging groups. Significant bilateral ventricular morphometric differences ( $p < 0.025$ ) were revealed on an Arizona APOE cohort, which includes 18 cognitively unimpaired adults progressing within two years to cognitive decline (CD), and 20 matched controls with at least 4 years of post-baseline cognitive stability (CS). The VMAS also detected significant differences in bilateral ventricular morphometry ( $p < 0.035$ ) in 44 Alzheimer's disease Neuroimaging Initiative (ADNI) subjects (18 CD and 26 CS) with the same inclusion criteria. Both sets of experimental results demonstrated that frontal horn regions on bilateral ventricles are affected in cognitively declining subjects, and the left ventricle has higher effect size than the right.

Conclusions: To our knowledge, this is the first work to detect ventricular abnormalities of normally aging subjects prior to cognitive decline compared to subjects that remain cognitively stable. The key finding of this work is that the VMAS promises to track disease progression at a subregional level and measure the effects of pharmacological intervention in the preclinical stage.

**PUTATIVE ROLES FOR NEUROFILAMENTS IN THE DEVELOPMENT OF ALZHEIMER'S DISEASE.** Day WA. University of Arizona; Arizona Alzheimer's Consortium.

Abstract: A putative role for neurofilaments, particularly the species migrating at 64kD (NFL), in the development of neurofibrillary tangles (NFTs) was suggested over 30 years ago. More recently, a direct role for NFL in maintaining synaptic integrity and (to an extent) calcium homeostasis by modulation of Glutamate Receptors has also been recognized. Of particular note, formation of NFTs, loss of synaptic integrity and calcium homeostasis are all features of Alzheimer Disease (AD) pathogenesis.

Early studies dealt with cleavage of neurofilament species by calcium-activated and serine-specific endogenous proteases, the original contention being that solubilization-resistant fragments are minor components of solubilization / re-aggregation purification. However, it has since been found that insoluble fragments are generated in large numbers over time, presumably by a "seeding" process, which is a feature of amyloid-related diseases, including Alzheimer's Disease. It has also been shown that blots of fragments migrating to 52kD and 58kD are labelled by anti-NFL antibody and that antibody raised against NFF58 fragments stimulate clumping of NFF52-58 material and label NFL filaments.

A direct involvement of NFL cleavage in AD pathogenesis, in addition to the accepted role as a marker, is thus indicated. That is, by direct effects of synaptic integrity and calcium homeostasis, but also on development of the neurofibrillary tangles which are a major feature of progression of Alzheimer's Disease.

**RNA STRESS GRANULE COMPONENTS ARE DYNAMICALLY EXPRESSED DURING AGING AND STRESS CONDITIONS IN RATS AND FRUIT FLIES.** Eck R, Chawla MK, Siddegowda BB, Carey NJ, Zempare M2,3, Nguyen C, Billheimer D, Barnes CA, Zarnescu DC. University of Arizona; Arizona Alzheimer's Consortium.

Background: During times of cellular insult, non-membrane bound organelles called RNA stress granules form to sequester mRNAs, translation initiation factors and proteins into dense cytoplasmic structures, halting translation. Dysfunction in the dynamic assembly and disassembly of RNA SGs has been linked mechanistically to age-related neurodegenerative diseases. It is not known, however, how these mechanisms are affected during aging.

Methods: To understand the molecular mechanisms that underly these links, we profiled the dynamic changes of RNA SGs during aging and their relationship to stress resiliency through examining the expression of genes critical to translation initiation and RNA SGs formation in behaviorally characterized young (6-10 mo, n=11), middle-aged (15-19 mo, n=11), and old rats (23-25 mo, n=13). These rats were assessed for their spatial memory, working memory, and motor function using the Morris water maze. Some of the genes profiled were G3BP1, necessary for RNA SGs formation, FMRP, a modulator of mRNA association with RNA SGs, EIF2alpha, a translation initiation factor whose phosphorylation indicates RNA SG formation, and PABP and TIAR among others. Western blots and real-time PCR found region-specific expression of these critical genes in the hippocampus, pre-frontal cortex, and cerebellum throughout aging. Using regression models, we sought to determine if these region-specific expression changes can account for variation among rats in behavioral performance, when taking age into account. We used Principle Component Analysis to determine if the variations among critical protein and transcript expression levels reveal differences or similarities between rats that relate to age or behavioral performance.

Results: The levels of key proteins and transcripts, along with other analysis trends, will be compared between young and old rats who received maximum electro-convulsive shock treatment (shock duration = 1 second, current intensity = 85mA, 1 hour recovery) and those who did not. In fruit flies, the dynamic response of RNA SGs under stress conditions appears to vary with age compared to the non-stressed control condition.

Conclusions: Future investigation will focus on isolating RNA SGs from fruit flies to examine how components associated with RNA/protein structures change during aging and in response to multiple stress conditions.

**CELL-SPECIFIC RNASEQ REVEALS GENDER-SPECIFIC GENE EXPRESSION CHANGES IN OLIGODENDROCYTES AND NEURONS IN ALZHEIMER'S DISEASE.** Enriquez D, Elyaderani A, Henson S, Antone J, Geiger P, Adkins JR, Tassone E, Serrano GE, Beach TG, Readhead B, Wang Q, Mastroeni D, Dudley J, Reiman EM, Liang WS. Translational Genomics Research Institute; Banner Sun Health Research Institute; Arizona State University; Icahn School of Medicine at Mt. Sinai; Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: There is a critical need to clarify molecular mechanisms involved in the development of Alzheimer's disease (AD), and use them to discover promising treatments. Detailed molecular data from specific brain cells and brain regions that are differentially affected by AD can inform experimental studies and clarify the extent to which their findings are relevant to this fundamentally human disease. The goal of this study is thus to generate a public resource to advance the scientific understanding, treatment, and prevention of AD.

Methods: To generate a transcriptomic reference, we are performing analyses on fresh frozen brain (median PMI ~3 hrs) from approximately 50 expired donors (including those with clinical and neuropathological evidence of dementia due to AD and donors without cognitive impairment or neuropathological criteria for AD [no disease, ND]). Analyses are focused on regions differentially impacted by AD, including the entorhinal cortex, hippocampus, posterior cingulate (PC), superior frontal gyrus (SFG), and primary visual cortex (VCX); and include: (1) laser capture microdissection (LCM) and total RNA sequencing (RNAseq) of neurons, astrocytes, and microglia; (2) bulk RNAseq; (3) single nuclei RNAseq (snRNAseq) for the PC, SFG, and VCX; and (4) whole genome sequencing.

Results: While analyses are ongoing, we present here snRNAseq (n=45) and non-tangle bearing neuron RNAseq (n=53) data generated from the SFG. Using snRNAseq, six major cell populations were identified across 32,507 high quality nuclei, including pyramidal and GABAergic neurons, astrocytes, microglia, oligodendrocytes (OD), and oligodendrocyte precursor cells. AD OD nuclei demonstrated increased expression (corrected  $P < 0.05$ ) of OD differentiation and specification genes including SOX8, SOX10, and OLIG1. These changes coincide with decreased expression of OD marker genes such as MBP and MOG, which encode factors required for myelin, in AD nuclei. SnRNAseq further revealed gender-specific neuron populations and increased expression of genes involved in developmental processes, including PAX6 and ARX, in AD male neurons compared to AD female neurons. Elevated expression of these genes was observed in LCMed non-tangle bearing neurons in AD males, although statistical significance was not reached. LCM RNAseq data, for which we generated a median of >124 million total reads across all samples, also revealed more gender-associated gene expression changes (corrected  $P < 0.05$ ) in the context of AD compared to disease-specific changes (AD vs ND) alone.

Conclusions: Our findings corroborate previous studies that describe gender-specific brain differences in AD, as well as studies that have reported deficiencies in myelination processes, and white matter changes, in AD. As we continue to generate data, we will assess if observed changes extrapolate across other brain regions. In addition to generating this resource for the research community, we will construct a multi-scale network of AD to identify novel disease drivers and therapeutic targets in order to develop improved treatments for patients.

**AD-NET: AGE-ADJUST NEURAL NETWORK FOR IMPROVED MCI TO AD CONVERSION PREDICTION.** Gao F, Yoon H, Xu Y, Goradia D, Luo J, Wu T, Su Y. Arizona State University; Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: The prediction of Mild Cognitive Impairment (MCI) patients who are at higher risk converting to Alzheimer Disease (AD) is critical for patient selection in clinical trials and effective disease management. Neuroimaging plays critical role in the understanding of AD and holds great promise to provide important disease markers for diagnosis and prognosis of AD including assessing the conversion risks. While Deep learning (DL) techniques may further enhance the utility of neuroimaging in the diagnosis and prognosis of AD, one great challenge facing deep learning is the limited imaging data available. To tackle this issue, transfer learning (TL) has been investigated to address this limitation and extend the applicability of deep models. In this work, we examine the utility of brain structural age derived from deep models of T1-weighted MR images in the prediction of MCI participants in the ADNI cohort who are at higher risks of converting to AD.

Methods: We propose an AD-NET (Age-adjust neural network) that is constructed by two parallel 3D-convolutional neural network (CNN) one for age prediction and the other optimally fused the predicted age as a surrogate marker to assist the MCI to AD conversion risk prediction. The two networks were combined in the final layer to determine the final estimation of the conversion risk with the integration of chronological age as an independent input. The 3D-CNN was trained for age estimation using a pooled dataset of 847 cognitively normal participants (aged 18-94 yrs) from two database (IXI and ADNI). The 3D-CNN model and its weights were transferred to the second stage of training to optimize the age prediction in MCI cohort and conversion risk prediction. This second stage of training and cross-validation was performed based on 297 MCI participants in ADNI among them 168 converted to AD within 3 years. 5-fold cross-validation was performed and the performance of the conversion risk prediction task was compared with other commonly used techniques.

Results: The proposed AD-NET approach achieved a sensitivity of 0.80 and specificity of 0.73 and an area under the ROC curve of 0.77 in the prediction of 3-year MCI to AD conversion risk. In comparison, a deep polynomial network combined with a support vector machine technique achieved a sensitivity of 0.68 and a specificity of 0.87 which used both structural MR and amyloid PET images.

Conclusions: Our proposed AD-NET achieved similar performance in the prediction of MCI to AD conversion risk as state-of-the-art techniques which often requires more biomarkers than structural MR. Further investigation is ongoing to improve the AD-NET technique.

**PET EVIDENCE OF PRECLINICAL CEREBELLAR AMYLOID PLAQUE DEPOSITION IN AUTOSOMAL DOMINANT ALZHEIMER'S DISEASE.** Ghisays V, Lopera F, Goradia DD, Protas HD, Malek-Ahmadi MH, Chen Y, Devadas V, Luo J, Lee W, Brown CT, Baena A, Bocanegra Y, Guzman-Velez E, Paredilla-Delgado E, Vila-Castelar C, Fuller JT, Hu N, Clayton D, Smith J, Thomas RG, Toga AW, Alvarez S, Rios-Romenets S, Langbaum JB, Chen K, Su Y, Tariot PN, Quiroz YT, Reiman EM, API ADAD Colombia Trial Group. Banner Alzheimer's Institute; Universidad de Antioquia, Medellin, Colombia; Massachusetts General Hospital, Harvard Medical School; Boston University; Genentech Inc.; Roche Products Ltd.; University of California San Diego; University of Southern California; Hospital Pablo Tobon Uribe, Medellin, Colombia; Arizona State University; University of Arizona; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

**Background:** In contrast to late onset Alzheimer's disease (LOAD), the late clinical stages of autosomal dominant Alzheimer's disease (ADAD) are associated with greater neuropathological evidence of cerebellar amyloid plaque (A $\beta$ ) deposition. In this study, we used PET measurements of fibrillar A $\beta$  burden to characterize the presence and age at onset of cerebellar A $\beta$  deposition in cognitively unimpaired Presenilin-1 (PSEN1) E280A mutation carriers from the world's largest extended family with ADAD.

**Methods:** Florbetapir and PiB PET data from two independent studies – API ADAD Colombia Trial (NCT01998841) and the Colombia-Boston (COLBOS) longitudinal biomarker study were included. Template-based cerebellar standard uptake value ratios (SUVR), using a known-to-be-spared pontine reference region, were used to 1) compare 290 cognitively unimpaired 28-56 year-old mutation carriers and non-carriers; 2) characterize associations among cerebellar-to-pontine SUVR and age, mean cortical SUVRs, and episodic memory performance; and 3) estimate the age at which cerebellar-to-pontine SUVRs begins to differ significantly in the carrier and non-carrier groups.

**Results:** Compared to non-carriers, cognitively unimpaired carriers had higher cerebellar-to-pontine florbetapir and PiB SUVRs ( $p < .0001$ ). Mean cortical SUVRs were positively correlated with age, and negatively correlated with episodic memory performance ( $p < .05$ ). SUVRs began to distinguish carriers from non-carriers at age 34, 10 years before the carriers' estimated age at mild cognitive impairment onset.

**Conclusions:** This PET study provides evidence of cerebellar A $\beta$  plaque deposition in cognitively unimpaired mutation carriers starting years before their clinical onset. Additional studies are needed to clarify the impact of using a cerebellar versus pontine reference region on the power to detect and track ADAD progression, even in preclinical stages of this disorder.

**FEASIBILITY OF ULTRASOUND-ASSISTED LUMBAR PUNCTURE IN COGNITIVE AGING AND ALZHEIMER'S DISEASE AND RELATED DISORDERS RESEARCH.** Goldfarb D, Viramontes D, Callan M, Liebsack C, Goddard M, Reade M, Weidman D, Tsai P, Atri A. Banner Sun Health Research Institute; Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

**Background:** Precise planning, accurate anatomical site selection, appropriate execution, and making individualized and dynamic intra-procedure adjustments are crucial to achieving a successful lumbar puncture (LP) for collection of cerebrospinal fluid (CSF). These factors are particularly germane when performing LPs in older individuals or those with neurodegenerative cognitive-behavioral/motor diseases in the Alzheimer's disease and related disorders (ADRD) spectrum. In cognitive aging and ADRD-related research with predominantly elderly individuals several technical and non-technical factors, including preconceived perceptions and age-, disease- and illness-related factors, can contribute to the challenges and success a LP. Ultrasound-assisted lumbar puncture (Us-LP) has not been previously studied in ADRD-related research. This study aimed to assess the feasibility of training and implementing, and to explore the utility of, Us-LP by LP clinicians conducting aging- and ADRD-related research.

**Methods:** LP clinician-researchers from two Arizona ADRD centers completed simulation-based Us-LP training. LP clinician-researchers then had the option to use ultrasound assistance during scheduled LPs for ADRD-related research participants, who were consecutively enrolled. Prior to LP, limited demographic data was obtained, and after each LP clinician-researchers completed a questionnaire to assess LP details, use of ultrasound and rationale, and LP performance. Soon after the procedure, participants were contacted at home to assess adverse events (AEs). The study was conducted, with IRB approval, at the Banner Sun Health Research Institute in Sun City, and the Banner Alzheimer's Institute in Phoenix, AZ.

**Results:** Two expert ultrasound faculty trained six LP clinician-researchers (five neurologists and one advanced practice nurse) on Us-LP techniques using simulation-based models with the Philips Lumify system, a portable hand-held transducer which connects to a tablet. Images were obtained with the Lumify app (Philips Lumify, [usa.philips.com](http://usa.philips.com)). Between August 2019-March 2020 (when LPs were paused due to COVID-19 restrictions), 58 LP research participants enrolled, 66% female, mean age 71.4 years (SD 9.29). LP clinician-researchers implemented Us-LP into their LP practices and utilized Us-LP on 37/58 (64%) participants. In these 37, the following reasons for ultrasound use were reported: clinician preference 15/37 (41%), body habitus/difficult to palpate landmarks 8/37 (22%), chronic spine deformity 2/37 (5%), prior traumatic tap 1/37 (3%), and other 11/37 (30%).

**Conclusions:** It is feasible to train and implement portable hand-held Us-LP by clinician-researchers to collect CSF from older ADRD research participants. Preliminary analysis of pilot data suggests Us-LP may be of utility to ADRD clinician-researchers collecting CSF. Ongoing analyses and further data will be needed to determine whether availability and proficiency, and what factors, may contribute to Us-LP utility that could lead to greater clinician-researcher and participant confidence, higher success, better tolerability and decreased AEs for LPs. Improving these factors will aid to accelerate CSF biomarker-, aging- and ADRD-related research.

We thank the research participants for their generosity and altruism to make this research possible. This study was also made possible through funding by a 2019-2020 Arizona Alzheimer's Disease Consortium pilot grant (PI Goldfarb), funds from The BSHRI Sun Health Foundation Fund, and ultrasound equipment (Philips Lumify systems) provided by Philips Healthcare, a division of Philips Research North America LLC.

**GENEMATCH: SUCCESS OF REFERRING PARTICIPANTS TO THE GENERATION PROGRAM AND LESSONS LEARNED FOR FUTURE ENROLLMENT INTO ALZHEIMER'S DISEASE PREVENTION TRIALS.** Gordon D, Salata H, Walsh T, High N, Kettenhoven C, Reiman EM, Tariot PN, Langbaum JB. Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: We developed GeneMatch in 2015 to assist enrollment into Alzheimer's disease (AD) prevention studies by enriching referrals based in part on APOE4 genotype. The API Generation Program, which enrolled cognitively unimpaired (CU) APOE4 carriers ages 60-75 was the first multisite clinical trial to use GeneMatch as a recruitment source.

Methods: GeneMatch is a US-based, trial-independent recruitment program of the Alzheimer's Prevention Registry (APR), conducting APOE testing in CU individuals aged 55-75. GeneMatch does not disclose APOE results to participants directly, however, results are used to match participants to enrolling studies in a manner that does not inadvertently disclose genotype. Participants are notified by email when they have been invited to a study. A reminder email is sent 7 days later, a letter is mailed to the participant's home 2mo after if no response is received. Once a participant accepts his/her invitation, their contact information is shared with their preferred study site through an online portal. In July 2016, GeneMatch began inviting participants to the Generation Program.

Results: To date, >90,000 people have joined GeneMatch, > 30% are APOE4 carriers. Recruitment in the Generation Program and treatment with umibecestat was terminated in July 2019 after an early signal of mild worsening in some measures of cognitive function. At that time, 16,521 GeneMatch participants had been invited to the Generation Program, 33% (5,461) of whom accepted their invitations. Of those who accepted, 2,202 consented to the Generation Program (randomization data not yet available), 292 were scheduled for screening, 511 were awaiting scheduling for screening, 242 were waiting to be contacted by their selected study site, 1,209 were found to be ineligible after a phone screen by sites, 695 were no longer interested, and 310 were lost to follow-up.

Conclusions: The Generation Program was on track to complete enrollment in 2019 and GeneMatch was a successful program in helping to recruit APOE4 carriers. Lessons learned include the importance for GeneMatch to coordinate with study sites to ensure they are prepared to receive referrals Future analyses will examine success of GeneMatch (vs. other recruitment sources) for randomization.

**PERINEURONAL NETS IN THE CEREBRAL CORTEX OF COGNITIVELY-ASSESSED AGED MACAQUE MONKEYS.** Gray DT, Pyon W, De La Pena NM, Schwyhart R, Wallace E, Puchta J, Hartig W, Barnes CA. University of Arizona; University of Leipzig, Germany; Arizona Alzheimer's Consortium.

Background: Perineuronal nets (PNNs) are specialized extracellular matrix structures that envelop specific neurons in the central nervous system and play critical roles in controlling plasticity and maintaining synaptic function (Sorg et al., 2016, J Neurosci). Alterations in the expression of different components of the extracellular matrix have been shown to occur in normative brain aging, as well as in several nervous system disorders. No studies have investigated PNNs across the lifespan of behaviorally characterized, aged nonhuman primates. Furthermore, the impact that potentially altered PNNs have on the manifestation of different aspects of age-associated cognitive decline is not clear.

Methods: To these ends, the present study used fluorescence labeling and unbiased quantification of perineuronal net markers [Wisteria floribunda agglutinin (WFA) and the chondroitin sulfate proteoglycan aggrecan] on the brains from a colony of 30 rhesus macaque monkeys ranging in age from 7 to 32 years. All of these monkeys also underwent tests of spatial short-term memory (delayed response), object recognition memory (delayed nonmatching-to-sample), and object discrimination, which allowed relationships between PNNs and cognition to be investigated.

Results: While there are interesting trends with respect to age, PNNs and parvalbumin (PV)-immunoreactive neurons, our preliminary results (N = 3 aged, mean 28 years; N = 3 adult, mean 11 years) suggest no age effect. The data do suggest that the strongest associations are found between the proportion of PV-immunopositive neurons with nets and behavior. Specifically, animals with more perineuronal nets surrounding PV-immunoreactive neurons tended to show worse behavior on all of our cognitive tasks. Furthermore, animals that exhibited fewer PNNs associated with PV-immunopositive cells tended to show better behavioral performance on our tasks.

Conclusions: We are currently expanding the sample size, PNN markers, and the brain regions analyzed to more thoroughly characterize these nets across aging.

**CLINICOPATHOLOGICAL CORRELATION: DOPAMINE AND AMYLOID PET IMAGING WITH NEUROPATHOLOGY IN THREE SUBJECTS CLINICALLY DIAGNOSED WITH ALZHEIMER'S DISEASE OR DEMENTIA WITH LEWY BODIES.** Gupta HV, Beach TG, Mehta SH, Shill HA, Driver-Dunckley E, Sabbagh MN, Belden CM, Liebsack C, Serrano GE, Sue LI, Pontecorvo MJ, Mintun MA, Joshi AD, Adler CH. University of Kansas Health System; Banner Sun Health Research Institute; Mayo Clinic College of Medicine; Barrow Neurological Institute; Ruovo Clinic; Avid Radiopharmaceuticals; Arizona Alzheimer's Consortium.

Background: Imaging biomarkers have the potential to distinguish between different brain pathologies based on the type of ligand used with PET. AV-45 PET (florbetapir) is selective for the amyloid plaques of Alzheimer's disease (AD) while AV-133 PET is selective for VMAT2, a dopaminergic marker depleted in Parkinson's disease (PD) and dementia with Lewy bodies (DLB). The objective of this study was to report the clinical, AV-133 PET, AV-45 PET, and neuropathological findings of three clinically diagnosed dementia patients who were part of the Avid Radiopharmaceuticals AV133-B03 study as well as the Arizona Study of Aging and Neurodegenerative Disorders (AZSAND).

Methods: Subjects were recruited for the AV133-B03 study, were assessed by neuropsychologists and neurologists, died and had autopsy with neuropathological examination. A total of 3 subjects who had PET imaging with both AV-133 and AV-45 as well as a standardized neuropathological assessment were included. The final clinical, PET scan, and neuropathological diagnoses were compared.

Results: The first subject had a clinical diagnosis of dementia with Lewy bodies (DLB). His AV-133 PET showed bilateral dopaminergic degeneration and AV-45 PET was positive for amyloid. The final diagnosis based on clinical and pathological information was DLB and AD. The second subject was diagnosed clinically with probable AD and AV-45 PET was positive for amyloid while AV-133 PET was normal. Neuropathological diagnostic criteria were met for both DLB and AD. The third subject had a clinical diagnosis of DLB. Her AV-45 PET was positive for amyloid and AV-133 showed dopaminergic degeneration. The final diagnosis based on clinical information and pathology was multiple system atrophy (MSA) and AD.

Conclusions: PET imaging using AV-133 for the measurement of VMAT2 density can help distinguish between AD and DLB. However, some cases of DLB with less-pronounced nigrostriatal dopaminergic neuronal loss can potentially be missed.

**THE ALZHEIMER'S PREVENTION REGISTRY: ACCELERATING RECRUITMENT AND ENROLLMENT INTO ALZHEIMER'S-FOCUSED STUDIES.** High N, Kettenhoven C, Graf H, Reiman EM, Tariot PN, Langbaum JB. Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: The Alzheimer's Prevention Initiative (API) is a collaborative program conducting preclinical Alzheimer's disease (AD) trials in people who are at elevated risk of developing AD symptoms. To support these and other trials, we developed the web-based Alzheimer's Prevention Registry ("APR") program to provide resources to the AD scientific community, aiming to accelerate recruitment and enrollment into studies and complement local efforts.

Methods: Adults age 18+ are eligible to join APR at [www.endALZnow.org](http://www.endALZnow.org). At enrollment, individuals provide their name, email address, zip/postal code and year of birth; after enrollment they provide additional demographic information at their convenience with the option to select "prefer not to answer." The APR team works with researchers to promote AD-focused studies to members via its website and email. APR members receive email communication for engagement purposes and are notified when new study opportunities are available. Members interested in a study fill out an online contact form with their names, email addresses and phone numbers, review and acknowledge the study's eligibility criteria and authorization for APR to share their contact information with the enrolling study team. The APR team provides enrolling studies with a secure dashboard for tracking referrals.

Results: The APR launched in May 2012; as of January 2020, >347,000 have joined. APR members have a mean age of 63.3 years (SD 11.7), are predominately female (75%), self-report being cognitively unimpaired (94%), and 50% report a family history of AD/dementia. To date, the APR has promoted 82 AD-focused studies, results from which will be presented. Only anecdotal enrollment metrics are available prior to the launch of the online contact form, however web-traffic metrics that are available from our promotions of online studies (e.g., TRC-PAD, Brain Health Registry) suggest >16,000 APR members expressed interest in joining these studies. The 6-month pilot phase of the online contact form included 7 studies and referred 320 APR members.

Conclusions: Enrollment into the APR continues to increase, as does the number of studies the APR promotes to its members. We are exploring novel approaches for increasing enrollment and engagement of members, and collaborating with researchers and sponsors to help promote studies in their communities.

**EARLY DYSFUNCTIONAL MOLECULAR PATHWAYS OF ALZHEIMER'S DISEASE IDENTIFIED BY A NOVEL SINGULAR VALUE DECOMPOSITION ALGORITHM.** Huseby CJ, Delvoux E, Coleman PD. Arizona State University; Arizona Alzheimer's Consortium.

Background: Microarray technology is an effective tool to capture gene expression of thousands of genes from multiple molecular pathways in human brain tissue. Many expression datasets exist holding information about differences in gene expression related to Alzheimer's disease processes. Often research groups employ comparative analysis strategies on the data resulting in differential expression between disease and controls. However, expression may not necessarily increase or decrease with disease progression but have highly variable patterns indicating a dysregulation in cellular homeostasis functions.

Methods: Using available expression datasets with multiple brain regions as a function of Braak-staging, we incorporate a variation on singular value decomposition (SVD) as a rigorous approach to look for expression signatures that have highly variable patterns indicating a molecular process dysregulation. By pathway analysis, we determined upstream players in the affected biological pathways of the identified transcripts and will proceed to design appropriate experiments to validate the early dysregulated molecular pathways.

Results: Analysis of the sequential singular values generated determines the number of components that capture the data essential features. The magnitude of the singular values identified dysfunctional expressed transcripts at early stage in the disease before the appearance of tau protein neurofibrillary tangles (NFT). In the early stages, we find that brain regions already affected by NFT, show large expression variation in transcripts related to inflammation. In brain regions not yet affected by NFT, the biological processes that contain dysregulated transcripts include extracellular matrix organization, glycosaminoglycan metabolic processes, oxygen transport, etc.

Conclusions: Using a novel approach to analyze expression data with multiple brain regions and Braak-staging, we identified transcripts displaying dysregulated expression signatures at early stages of disease in brain regions not yet affected by neurofibrillary tangles. The relationships of these transcripts and pathways enriched guide us to explore upstream events in the early disease cascade.

**PLASMA AMYLOID-BETA42/40 RATIO AS BIOMARKER OF CEREBRAL AMYLOIDOSIS IN COGNITIVELY UNIMPAIRED APOE4 HOMOZYGOTES, HETEROZYGOTES AND NON-CARRIERS.** Jansen WJ, Ghisays V, DeMarco K, Boker C, Chen K, Chen Y, Luo J, Protas H, West T, Meyer M, Kirmess K, Verghese P, Hu H, Yarasheski K, Su Y, Reiman EM. Banner Alzheimer's Institute; C2N Diagnostics; Arizona Alzheimer's Consortium.

**Background:** Plasma amyloid- $\beta$  (A $\beta$ ) blood tests have great promise in research and care. C2N Diagnostics has developed a mass spectrometry-based plasma A $\beta$ 42/40 assay (APTUSTM-pA $\beta$ 42/40), which has previously demonstrated approximately 88% accuracy (81% sensitivity and 83% specificity; CTAD 2019) to distinguish between individuals with A $\beta$ -positive and negative (A+ and A-) PET scans. We sought to characterize the assay's ability to discriminate cognitively unimpaired APOE4 homozygotes, heterozygotes, and non-carriers from the Arizona APOE Cohort.

**Methods:** Plasma A $\beta$ 42/40 concentration ratios and APOE4 gene dose (0,1,2 alleles) were characterized blindly using 0.5 cc plasma aliquots from 121 cognitively unimpaired 66 $\pm$ 8 (47-85) year-old participants, including 21 APOE4 homozygotes, 37 heterozygotes and 63 non-carriers whose blood samples and PiB or florbetapir PET scans were acquired at the same visit. Neuropathologically validated PiB 1.47 and florbetapir 1.17 SUVR cut-offs were used to determine A+ and A- PET scans. Receiver operating characteristic (ROC) analyses were performed calculating the areas-under-the curve (AUCs) before and after adjustment for APOE4 gene status or dose, age and sex. The optimal A $\beta$ 42/40 cut-off, sensitivity and specificity were retrospectively characterized.

**Results:** Plasma A $\beta$ 42/40 ratios discriminated between individuals with A+ and A- PET scans with 0.87 and 0.89 AUCs, respectively, before and after adjustment for APOE4 status. Adjustment for APOE4 gene dose, age and sex did not significantly influence these results. In the APOE4 status-adjusted ROC, a plasma A $\beta$ 42/40 0.096 cutoff, with 91% sensitivity and 82% specificity had the maximum Youden Index. We observed trends of a greater percentage of A- PET APOE4 homozygotes than heterozygotes or non-carriers being plasma A+ (29%, vs. 11%; p=0.08, and vs. 11%; p=0.05, respectively). Further, individuals with an A+ plasma test but an A- PET scan were at 9-fold increased risk of conversion to A+ PET compared to individuals with an A- plasma test.

**Conclusions:** APOE4-adjusted plasma  $\beta$ 42/40 assays have great promise in AD research, prevention trials and clinical care, with demonstrated high accuracy for cognitively unimpaired persons with two, one or no copies of the APOE4 allele. This study suggests that an A+ plasma test result predicts progression from an A- to an A+ PET scan in the near future.

**A BRAIN TISSUE METABOLOMIC SIGNATURE DISCLOSES ALZHEIMER'S DISEASE POST-MORTEM.** Jasbi P, Shi X, Turner C, Eghlimi R, Jentarra G, Gu H. Arizona State University; Midwestern University; Arizona Alzheimer's Consortium.

**Background:** Alzheimer's disease (AD) is the most common cause of dementia, accounting for an estimated 60 to 80% of cases, and is the sixth-leading cause of death in the United States. A major challenge persists in the post-mortem diagnosis of AD patients from those suffering mild cognitive impairment (MCI) and control patients with high pathology (HPC). In this study, we aim to use mass spectrometry (MS)-based metabolomics, in conjunction with advanced multivariate statistics, to discover metabolic signatures of AD, which has high potential for disease diagnosis and therapy development.

**Methods:** In this study, we present a combination of gas chromatography-MS (GC-MS) for global metabolic profiling and analysis of long- and short-chain fatty acids, in addition to liquid chromatography-tandem MS (LC-MS/MS) for the detection of targeted aqueous metabolites and lipids. Samples were taken from four groups of subjects: 12 normal control (NC) patients, 12 subjects characterized as HPC, 12 with sub-clinical MCI, and 12 diagnosed with AD.

**Results:** Using this approach, 2084 metabolites and features from many relevant metabolic pathways of biological significance were reliably detected and monitored in 48 tissue samples harvested from the superior frontal gyrus of male and female subject's post-mortem. Multivariate significance testing informed the construction and cross-validation ( $p < 0.01$ ) of a partial least squares-discriminant analysis (PLS-DA) model defined by a 5-metabolite panel of potential diagnostic biomarkers. Receiver operating characteristic (ROC) analysis showed high predictive accuracy of the PLS-DA model for discrimination of NC (97.5%), HPC (95.0%), MCI (75.0%) and AD (93.1%). Pathway analysis revealed significant disturbances in lysine degradation, fatty acid metabolism, and the degradation of branched-chain amino acids. Over representation analysis of lipid signatures revealed significant alterations in glycerophospholipid and choline metabolism. Network analysis showed significant enrichment in 11 enzymes, mostly of the mitochondria.

**Conclusions:** The results expand basic knowledge of the metabolome related to AD pathogenesis and reveal pathways that can be targeted therapeutically in future studies. This study also provides a promising basis for the development of larger multi-site projects to validate these candidate markers in readily available biospecimens such as blood to enable the effective screening, rapid diagnosis, accurate surveillance, and therapeutic monitoring of AD across population groups and further improve clinical care for AD patients.

**CORRELATIONS OF MICROBIAL COMPONENTS AND HOST RESPONSE MOLECULES IN THE BRAIN TISSUE OF AD PATIENTS AND CONTROLS WITH INDICATORS OF SYSTEMIC IMMUNOLOGICAL ACTIVATION IN THE SAME SUBJECTS.** Jentarra G, Chu P, Elliott N, Hudson H, Jones TB, Potter P. Midwestern University; Arizona Alzheimer's Consortium.

**Background:** Much of the brain pathology characteristic of Alzheimer's disease (AD) suggests an infectious process. We previously performed 16S rRNA gene sequencing analysis of DNA from post-mortem brain tissue of AD patients and controls and found that all subjects had some level of bacterial DNA in their brain tissue. We followed this with analysis of brain tissue and serum from the same subjects for gram-negative (LPS) and gram-positive (LTA) bacterial products, which supported what was observed in the sequencing data. We also performed Western blot analysis of brain tissue for molecules involved in host response to infection. Finally, we assessed systemic immunological status by examining TNF-alpha production in spleen and C-reactive protein (CRP) production in liver from the same subjects. We then performed correlation studies to detect any relationships between those various pieces of data.

**Methods:** Tissues used were frozen post-mortem serum samples plus frozen matched brain, liver, and spleen samples acquired from the BSHRI Brain and Tissue Bank. There were four subject groups: normal controls, high pathology controls, patients with mild cognitive impairment (MCI), and AD patients. For all brain, liver, and spleen tissue samples, and for matched serum samples from MCI and AD patients, n = 12. However, serum was not available for all normal and high pathology controls (n = 8 for normal controls and n = 11 for high pathology controls). PBS and RIPA lysates of tissues were made for analysis of specific molecules by ELISA and Western blot.

**Results:** We previously presented data showing a lack of strong correlations specifically between brain LPS/LTA levels and subject characteristics (age, sex, post-mortem interval (PMI), MMSE, APOE status, plaques, Braak score, and CAA). Those brain levels also did not show an association with general infections and inflammatory conditions present in patients and controls at the time of death, or with serum LPS levels, indicating that LPS levels in brain versus periphery are disconnected. Here, we expand our analysis of correlations to include evaluation of the 16S rRNA gene identification of bacteria versus brain LPS and LTA levels. We also assess the relationship of all data to brain fibrinogen and ferritin levels, which are frequently elevated in AD and can be indicators of host immune response to microbes. Additional correlations evaluate the relationship between factors present in the brain and systemic immunological activation indicators including TNF-alpha levels from spleen samples and CRP levels from liver samples.

**Conclusions:** Cumulatively, 16S rRNA gene sequencing data and brain LPS/LTA levels support the presence of bacteria, or at least bacteria-associated molecules, in the brain tissue of both AD patients and controls. This has led to the interesting hypothesis that the presence of some level of bacteria in the brain is likely to be normal in everyone. The correlations performed here are an attempt to determine if that microbial population can induce local or systemic effects that are associated with the development of Alzheimer's disease.

**ASSOCIATION BETWEEN CEREBRAL ARTERIOLE MEDIN AND PATHOLOGIC FEATURES OF VASCULAR DEMENTIA AND ALZHEIMER'S DISEASE.** Karamanova N, Truran S, Serrano GE, Hansen M, Davies HA, Madine J, Beach TG, Migrino RQ. Phoenix Veterans Affairs Health Care System; Banner Sun Health Research Institute; University of Liverpool; University of Arizona College of Medicine-Phoenix; Arizona Alzheimer's Consortium.

**Background:** Medin, a 50 amino acid amyloidogenic peptide, is one of the most common human amyloid proteins that accumulate in the vasculature with aging. We showed that medin induced pro-inflammatory activation in endothelial cells (ECs) and endothelial dysfunction in human brain donor collateral cerebral arteries. In-vitro, we also showed that medin-treated ECs resulted in astrocyte activation. These findings suggest that medin may play a role in cerebrovascular inflammation and dysfunction that may influence neuroinflammation and neurodegeneration, features found in vascular dementia (VaD) and Alzheimer's disease (AD). Our aims are to determine if cerebral arteriole medin is 1) higher in VaD or AD compared to non-demented (ND) controls and 2) associated with pathologic features of VaD or AD.

**Methods:** From the Sun Health Research Institute Brain and Body donation program, middle frontal gyrus histologic sections from 16 ND, 26 VaD and 33 AD donors were stained with anti-medin antibody (DAB staining) and medin content in each visualized arteriole was quantified using a scale of 0 (none) to 3 (extensive and abundant) by 2 independent reviewers blind as to diagnosis, and the values averaged for all the arterioles for each subject. Arteriole medin scores were compared among ND, VaD and AD donors, and correlated with final brain pathologic measurements of total plaque, plaque density score, total tangle, cerebral amyloid angiopathy (CAA) score, total white matter lesion score, total microinfarct score, lacunar infarct score and circle of Willis (CoW) pathology score. Multiple regression analysis was performed to determine the independent association of vascular measures of arteriole medin, CoW and CAA scores and age to total tangle, plaque density and total white matter lesion scores.

**Results:** Compared to ND, VaD and AD donors had higher cerebral arteriole medin scores ( $1.16 \pm 0.2$ ,  $1.84 \pm 0.1$  and  $2.0 \pm 0.1$  AU, respectively,  $p < 0.001$  VaD vs. ND and AD vs. ND). Arteriole medin scores showed significant correlations with total plaque ( $R = 0.34$ ,  $p = 0.008$ ), plaque density score ( $R = 0.32$ ,  $p = 0.01$ ), total tangle ( $R = 0.46$ ,  $p < 0.001$ ) and white matter lesion score ( $R = 0.28$ ,  $p = 0.03$ ), but not with CAA, CoW, microinfarct or lacunar infarct scores. In regression analyses, arteriole medin was independently associated with total tangle, plaque density and white matter lesion scores.

**Conclusions:** Cerebral arteriole medin is higher in VaD and AD compared to ND controls and is independently associated with pathologic lesions of AD and VaD. Further investigation is needed to assess whether medin plays a pathologic role in VaD and AD. In the context of prior observations of medin's in vitro effects on vascular cells, medin may be a novel candidate risk factor and potential treatment target for VaD and AD.

**THE ALZHEIMER'S PREVENTION REGISTRY: SUCCESS METRICS AND LESSONS LEARNED IN REGISTRY MEMBER RETENTION THROUGH ONLINE ENGAGEMENT CAMPAIGNS.** Kettenhoven CL, High NM, Salata HP, Reiman EM, Tariot PN, Langbaum JB. Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

Background: We developed the web-based Alzheimer's Prevention Registry (APR) program to provide resources to the AD scientific community, aiming to accelerate recruitment and enrollment into studies and to complement local efforts. Considerable efforts have been given to strengthening retention and engagement of APR members over time, regardless of whether they enroll in an AD study.

Methods: Adults age 18+ are eligible to join the APR at [www.endALZnow.org](http://www.endALZnow.org). At enrollment, individuals provide their name, email address, zip/postal code and year of birth; after enrollment they provide additional demographic information. Immediately after enrollment members begin receiving APR emails welcoming and orienting them to the APR. The APR sends members monthly newsletters and study opportunity announcements via email for engagement and retention purposes. We developed a 4-part re-engagement campaign for members who either have not opened an APR email within 6 months or fall below our engagement scoring model. If a member does not act on a re-engagement email, then APR removes them from its email distribution list. Only "actively engaged" members remain on the APR email distribution list.

Results: The APR has enrolled >347,000 members since its launch in 2012. As of January 2020, 86,175 people are considered "actively engaged" members of the APR. In 2019, 12 monthly e-newsletters were sent to APR members. The average e-newsletter open rate was 45% (compared to nonprofit healthcare industry average of 16%); and the average e-newsletter click rate (percentage of APR members who clicked on the email in relation to those who opened/viewed the email) was 24% (compared to the industry average of 1.6%). In 2019, 36 study opportunity emails were sent to APR members with similar open and click rates. To date, approximately 54,000 members have been added to the re-engagement campaign, and nearly 10,000 (18.5%) have been successfully re-engaged.

Conclusions: Deploying online engagement campaigns as a retention tool is a valuable resource for the APR. Thoughtfully engaging and re-engaging APR members promotes increased likelihood of email opens and clicks when study opportunities are offered. Future work will examine the relationship between email engagement and responsiveness to study opportunities.

**NEURAL CORRELATES OF EXPLORATION EXPLOITATION IN YOUNGER AND OLDER ADULTS EVENT-BASED FMRI.** Keung W, Wilson RC. University of Arizona; Arizona Alzheimer's Consortium.

Background: Throughout our lives, we often face decisions in which we must trade off the relative benefits of exploring options that are unknown and exploiting options we know well. As we age, such explore-exploit choices take on increasing importance as we decide how to invest savings in retirement (e.g. explore a new balance of stocks and bonds or exploit the balance we've used so far?), or how to treat a chronic disease (e.g. explore new treatments with unknown side effects, or exploit known treatments we've used for years?). Recently we have shown that young adults make explore-exploit decisions using a mixture of two strategies: exploration driven by information seeking, and exploration driven by adaptive behavioral variability. However, almost nothing is known about how these strategies, or the brain circuits that support them, change in old age.

Methods: Here we use a previously published task, the Horizon Task, to measure explore-exploit behavior. One important difficulty is that a participant's baseline level of exploration, such as behavioral variability driven simply by boredom or disengagement, is often indistinguishable from purposeful exploration. The key manipulation in the Horizon Task is the number of choices participants will make in the future — the time horizon. The idea behind this manipulation is that the tendency to purposefully explore should increase with horizon, while the baseline level of exploration should not. Thus, by comparing behavior between short and long horizons, exploration can be identified as the changes in information seeking and behavioral variability that occur with horizon. We scanned 30 younger adults and 17 older adults while they were performing this task.

Results: We found that in both younger and older adults, dorsal anterior cingulate cortex (dACC) and posterior cingulate cortex (PCC), show greater activation in long horizon than in short horizon, suggesting a possible role in promoting exploration. Conversely, inferior frontal gyrus (IFG) shows increased activation in short horizon relative to long horizon, suggesting a possible role in driving exploitation. Importantly, we also found that, while younger adults show increased BOLD activity in right IFG in short vs long horizon, older adults show bilateral IFG activation for the same contrast such that left IFG is significantly more active in older adults than younger adults.

Conclusions: Our results suggest that exploration and exploitation are implemented through dissociable brain circuits in both younger and older adults. Specifically, older adults recruit bilateral brain areas for exploitation, which is in line with previous evidence that healthy aging has been associated with switching from unilateral activation in younger adults to bilateral activation in older adults.

**INVESTIGATING LONG-TERM COGNITIVE EFFECTS OF VARIATIONS IN GYNECOLOGICAL SURGERY DURING ADULTHOOD IN A RAT MODEL.** Koebele SV, Woner VE, Northup-Smith SN, Willeman MN, Strouse IM, Bulen HL, Schrier AR, DeNardo DF, Mayer LP, Dyer CA, Bimonte-Nelson HA. Arizona State University; SenesTech, Inc.; Arizona Alzheimer's Consortium.

Background: It is currently estimated that by age 60, about a third of US women will have experienced hysterectomy, or the surgical removal of the uterus. This surgery is often performed in adulthood, prior to the final menstrual period. Although some women undergo a complete ovariectomy in adulthood, the ovaries are retained in about half of hysterectomy cases to avoid premature or abrupt surgical menopause. Research has suggested that hysterectomy, with and without ovarian conservation, can increase the relative risk of developing dementia compared to age-matched reproductive-tract-intact women. This association has piqued interest into how the non-pregnant uterus, which is typically considered to be an endocrine target but otherwise a dormant organ, may influence non-reproductive functions including cognitive processes. Our laboratory recently reported that hysterectomy with ovarian conservation resulted in detrimental effects on spatial working memory in adult Fischer-344-CDF rats six weeks after surgery (Koebele et al., 2019). The current study aimed to extend these findings and investigate the longitudinal cognitive effects of hysterectomy in adulthood in order to elucidate whether the cognitive effects observed six weeks post-surgery were transient and reversed with time or the observed cognitive changes were the beginning of a long-term, more global cognitive impairment that extends beyond the established working memory findings.

Methods: Adult female rats received either sham, ovariectomy, hysterectomy, or ovariectomy-hysterectomy surgeries. Rats were divided into three cohorts. The Adult cohort was tested six weeks after surgery, when rats were approximately seven months old. The Middle-Aged cohort was tested seven months after surgery, when rats were approximately 12 months old. The Aged cohort was tested one year after surgery, when rats were approximately 18 months old. Each cohort was tested on a battery of cognitive tasks evaluating spatial working and reference memory, including the water radial-arm maze. Rats were euthanized following behavior collection, and blood, ovaries, uteruses, and brains were collected for future analysis.

Results: Analyses of water radial-arm maze data indicate that the working memory impairment observed in adult hysterectomy rats six weeks after surgery was also observed at both the middle-aged and aged time points during learning. These behavioral findings will be discussed in the context of hormone and ovarian follicle profiles for each time point. Of note, the other behavioral evaluations for each cohort are in the process of being quantified and analyzed, and thus are not presented here.

Conclusions: Hysterectomy with ovarian conservation in adulthood appears to have long-lasting detrimental effects on spatial working memory, in turn altering the trajectory of cognitive aging compared to controls and other variations in gynecological surgery. It is critical to better characterize how variations in gynecological surgery impact cognition throughout aging in order to enhance quality of life and promote healthy brain aging for at-risk women.

**A COMPUTATIONAL MODEL OF AGED HEAD DIRECTION NETWORK UPDATING IN THE PRESENCE OF SUDDEN SPATIAL CUE MISMATCH.** Lester AW, Kapellusch AJ, Barnes CA. University of Arizona; Arizona Alzheimer's Consortium.

Background: As with older adults, aged rats show pronounced impairments on a number of different spatial navigation tasks as well as a bias toward relying on self-motion (i.e., idiothetic) over environmental (i.e., allothetic) cue-based navigation strategies (Lester et al., 2017). Rosenzweig et al. (2003) found that, when exposed to conflicting allothetic and idiothetic feedback, aged rats were impaired in navigating to an allothetic cue-aligned goal location and the place cell networks of aged rats were delayed in realigning their firing fields to match the spatial information relayed by the allothetic cues.

Methods: The Instantaneous Cue Rotation (ICR) task used here requires animals to navigate to a reward location that is always aligned to the projected visual cues in the environment (Lester et al., 2018). We previously reported that, when young and aged rats were tested on the ICR task, allothetic cues were found to exert a less pronounced influence on the running behavior of aged rats following sudden cue rotation. The overall pattern for young rats, in contrast, suggested a reliable although incomplete control by allothetic cues, which may reflect a greater tendency for young animals to resolve conflicting allothetic-idiothetic feedback by integrating information from both. A continuous attractor neural network model was created to assess how a sudden rotation of visual cues may affect the spatial tuning of head direction cell networks and how the behavior of these networks may be altered in the presence of erroneous self-motion feedback (i.e., idiothetic error). The model incorporates a head direction (HD) and angular head velocity (AHV) network, the tuning of which depend on both angular movement and visual cue inputs.

Results: In the absence of any idiothetic error, the HD and AHV networks collectively undergo a gradual but reliable realignment of their directional firing after visual cue rotation. In contrast, the introduction of idiothetic errors either amplifies or diminishes visual cue control over HD-AHV alignment depending on the degree and direction of drift these errors induce in the network's directional firing. As a consequence, visual cues exert less reliable control over directional tuning and often lose control over the networks directional tuning following visual cue rotation.

Conclusions: Considered in the context of known age-related changes in vestibular function as well as deficits in self-motion perception and path integration, the findings from this computational model suggest a plausible mechanism that could contribute to impaired integration of conflicting spatial signals in aged spatial networks.

**COMBINED GENISTEIN DIET AND EXERCISE PREVENT BOTH WEIGHT GAIN AND DEVELOPMENT OF KEY MARKERS OF ALZHEIMER'S DISEASE IN HIGH FAT-HIGH SUCROSE-FED MALE MICE.** Li R, Babu JR, Broderick TL, St Aubin C, Fisher A, Al-Nakkash L. Auburn University; Midwestern University; Arizona Alzheimer's Consortium.

Background: Chronic consumption of a western diet (high fat with high sugar, HFHS) is associated with metabolic syndrome, insulin resistance, type 2 diabetes, cardiovascular disease, loss of bone mass, inflammation, cognitive decline and increased risk of developing neurodegenerative diseases like Alzheimer's (AD). Genistein is a naturally occurring isoflavone found in soy, known to improve insulin sensitivity, and provide anti-inflammatory and neuroprotective value. Similar benefits have also been associated with moderate exercise. The aim of this study was to determine whether dietary genistein (600 mg genistein/kg diet, Gen) or moderate exercise (Ex), or both (Gen+Ex) would reduce the obese-diabetic phenotype and also limit progression of AD pathology in HFHS-fed mice.

Methods: C57BL/6J mice (5 weeks old) were randomly assigned to one of the following groups (n=10/group): lean control, HFHS, HFHS+Gen, HFHS+Ex, and HFHS+Gen+Ex. The HFD consisted of 60% saturated fat, 20% carbohydrate, 20% protein. Drinking water contained sucrose and fructose. Moderate exercise comprised daily treadmill running for 150 minutes/week for 12 weeks.

Results: Body weight was reduced 12-18% ( $P<0.05$ ) with Ex or Gen and reduced 42% ( $P<0.05$ ) by Gen-Ex combined compared to HFHS. The following data were obtained via western blot expression of proteins in brain homogenates: (1) 4G8 (A $\beta$  deposition) was significantly increased by HFHS diet and subsequently prevented by Gen or Gen+Ex, (2) PHF (phosphorylated Tau) was significantly increased by HFHS diet and subsequently prevented by Gen+Ex, (3)  $\beta$ -secretase-1 (BACE1,  $\beta$ -site APP cleaving enzyme 1, an enzyme involved in regulation production of A $\beta$ ) was significantly subsequently decreased by all treatment groups, and (4) ADAM metallopeptidase domain 10 (ADAM10, an  $\alpha$ -secretase enzyme which prevents generation of A $\beta$ ) was significantly decreased by HFHS diet and subsequently increased by all treatment groups. Interestingly, expression of NF- $\kappa$ B in brain homogenate was significantly diminished Gen or Gen+Ex.

Conclusions: We conclude that genistein and exercise often in isolation, but mainly in combination have significant benefits to prevent etiology of AD markers in the brains of HFHS-fed male mice. These benefits are associated with improvements in body weight and inflammatory response.

Support: Midwestern-Arizona Alzheimer's Consortium.

## **A PILOT STUDY OF 16 SUBJECTS TO DETERMINE THE CORRELATIONS BETWEEN THE LEVELS OF PLASMA DISEASE MARKERS AND THE SEVERITY OF BRAIN PATHOLOGY.**

Lue L-F, Serrano G, Chen W-P, Yang SY, Beach TG. Banner Sun Health Research Institute, MagQu LLC; MagQu Co. Ltd; Arizona Alzheimer's Consortium.

Background: Plasma biomarkers for neurodegenerative diseases have a potential role to be used as low-cost pre-clinical or clinical tools for assessing development, presence, or progression of the diseases. However, the success of developing such tools faced two challenges. First, the detection assays must be ultra-sensitive as many disease-specific markers are present at low levels in plasma. Second, the detected differences in plasma samples will need to accurately reflect the severity of the pathology in the brain, as determined by neuropathological examination. In this pilot study, we sought to address these challenges by using antemortem plasma samples collected close to the death of the patients (<1.5 years) and an ultra-sensitive ImmunoMagnetic Reduction (IMR) technology to detect several biochemical molecules relevant to neurodegeneration. Preliminary findings are reported here.

Methods: Plasma samples were separated from the blood drawn 18 months or less prior to death of donors enrolled at Brain and Body Donation Program (BBDP) at Banner Sun Health Research Institute. Samples used for the study had freezer storage duration shorter than 1.5 years. The study included 16 patients with absence or presence of various pathology, for examples: with the pathology characteristic of Alzheimer's disease, Parkinson's disease, Dementia with Lewy body (DLB), Fronto-temporal lobar degeneration (FTD), multi-system atrophy (MSA), Progressive Supranuclear Palsy (PSP), and mixed vascular dementia. A standard set of histology and histochemistry staining procedures and assessment were carried out on formalin-fixed brain tissues. Additional immunochemical characterization of the disease markers other than routine procedures were performed to detect pathological profiles of phosphorylated TDP-43 (p-TDP-43), Abeta 40 and Abeta 42 by the BBDP lab. Results were assessed semi-quantitatively. IMR assays were performed by MagQu LLT (Surprise, AZ, USA) to analyze the levels of Abeta40, Abeta42, total tau (t-tau), phosphorylated tau (p-tau), alpha synuclein (asyn), phosphorylated asyn (p-asyn), neurofilament light chain (NFL), and TDP-43. The patient serial number, demographic, clinical, histochemical, and neuropathological features were kept blinded at BBDP. After IMR data were sent to the BBDP, a compiled data file was then made available to the collaboration team for statistical analysis. The main objective of statistical analysis was to determine Spearman's correlations between the biochemical and pathological data.

Results: The ranges of expired age, years of education, and last MMSE and UPDRS scores of the cases were 65-96 years, 12-22 years, 10-30, and 0-40 respectively. The major findings from this study are: plasma p-asyn levels correlated with the summary neuropathologically-measured p-asyn ( $\rho=-0.555$ ,  $P=0.0255$ ) and regional densities in amygdala ( $\rho=-0.633$ ,  $P=0.0084$ ), cingulate cortex ( $\rho=-0.517$ ,  $P=0.0402$ ), and trans-entorhinal cortex ( $\rho=-0.519$ ,  $P=0.0395$ ). We also detected significant correlations between plasma TDP-43 levels and summary brain neurofibrillary tangle densities ( $\rho=-0.591$ ,  $P=0.0158$ ), as well as between plasma p-tau levels and neuropathologically determined phosphorylated TDP-43 density in amygdala ( $\rho=-0.409$ ,  $P=0.0495$ ).

Conclusions: The results of this pilot study showed that the levels of p-asyn, p-tau, and TDP-43 in plasma samples collected 1.5 years prior to death and measured by IMR assays were negatively associated with the densities of p-asyn, p-TDP-43, and neurofibrillary tangle pathology in postmortem brains. Future studies will determine the reproducibility of these results in a larger number of neuropathologically characterized cases

## **METHODS FOR POSTMORTEM HUMAN MICROGLIA BANKING AT HUMAN CELLS CORE FOR TRANSLATIONAL RESEARCH OF BANNER SUN HEALTH RESEARCH INSTITUTE.**

Lue L-F, Walker D, Beh S-T, Walker J, Sue LI, Serrano GE, Beach TG. Banner Sun Health Research Institute; Shiga Medical University; Arizona State University; Arizona Alzheimer's Consortium.

Background: Neuroinflammation is an important pathological feature in all neurodegenerative diseases. Although it is well known that microglia are the driving force of the inflammatory responses in aging and diseased brains, the delineation of the molecular mechanisms that are shared or specific to the diseases has been challenging. Current knowledge of microglia activation which is central to the inflammatory processes comes largely from research in mouse models. As immune responses in Alzheimer's transgenic mouse models cannot recapitulate completely the immune responses in human brains, there is a pressing need for using human microglia to bridge the gap. We have used human microglia isolated from brain tissues provided by the Brain and Body Donation Program (BBDP) of Banner Sun Health Research Institute for extensive research over the last two decades. Thus, we are in an advantageous position to use our expertise to build a human microglia banking program to meet the demand of human microglia for research. Our previous microglia processing was carried out immediately and continuously following brain removal from autopsies with short postmortem delay (< 5 hours). However, maintaining a 24-7 cell culture team is costly. Therefore, we have experimented with procedures to initially preserve cell viability, with later completion of cell processing.

Methods: At autopsy, frontal cortical brain tissues from BBDP donors were removed and immersed and refrigerated in Hibernate A media with antibiotics until processing. Processing consists of tissue dissection and enzymatic dissociation followed by separation of dead neurons, neuropil elements and red blood cells by gradient centrifugation. Remaining cells were washed and recovered by serial steps of centrifugation, resuspension, and filtration. Cells were then subjected to culture or cell-surface marker selection before cryoprotection or pelleting.

Results: We have tested a few modifications from our previous procedures, especially focusing on enzymes and media. Previously, papain digestion of brain tissues provided a good balance in microglia yield and cell types of interest (including harvest of brain microvascular endothelial cells and supporting cells such as smooth muscle cells and pericytes). However, papain is also known to strip off a range of cell-surface receptors affecting cell adherence and selection. Therefore, we experimented with tissue dissociation using enzymes such as dispase and collagenase. These enzymes exhibited a lower efficiency in dissociating elderly brain tissues compared to papain; we estimated 25-30% lower efficiency in 40-minute digestion time. Prolonged digestion marginally improved the efficiency. We also experimented with media enrichment, using supplement of B27 in Hibernate A media at the major steps of the isolation procedure, such as digestion and gradient centrifugation. We found that microglia exhibited healthier membrane morphology from this modification. We also performed antibody-conjugated magnetic bead selection of microglia from the initial glia cell mixture; we used pan microglia marker CD11b, resting microglia markers TMEM119, and inflammatory markers HLA-DR and TREM2. The yields of positively selected cells from the same marker varied between autopsy cases; within the same case, the yield of the positively selected cells varied between the markers.

Conclusions: We have incorporated enriched media to increase the success of microglia isolation in a two-stage process. We have optimized the selection procedure to allow separation of by markers representing different activation states.

**ANTICIPATED STIGMA AND DEMENTIA-RELATED ANXIETY IN MIDDLE-AGED AND OLDER ADULTS.** Maxfield M, Greenberg J. Arizona State University; University of Arizona; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease and related dementias (ARD) are common, and increasing age is the biggest risk factor (Alzheimer's Association, 2018). Early stages of dementia may require increased reminders and support to remain independent, but middle to late stages of dementia typically result in loss of independence, eventuating the need for continuous care for most, if not all, aspects of daily life. When considering the prevalence, negative perceptions of ARD, significant life changes, and lack of control associated with neurodegenerative disorders, it is perhaps unsurprising that ARD are a stigmatized and potentially anxiety-provoking set of diagnoses. The present study focused on the social consequences of ARD, specifically the anticipated stigma if one were to be diagnosed. Anticipated stigma involves concern about being treated differently based on one's diagnosis and originates from work concerning treatment of older adults' depression in primary care setting (Sirey et al., 2014). Older adults with depressive symptoms who anticipate stigma because of their diagnosis were less likely to receive mental health referrals (Sirey et al., 2014), and when enrolled in treatment, individuals anticipating greater stigma did not experience as much symptom reduction (Raeifar et al., 2017).

One may anticipate greater stigma when a diagnosis and its outcomes are perceived negatively, as in the case of dementia. Many negative stereotypes about older adults relate to impaired cognitive functioning. It was hypothesized that individuals anticipated greater stigma associated with ARD would also report greater dementia-related anxiety.

Methods: An online sample of 183 middle-aged and older adults completed self-report measures of dementia-related anxiety, anticipated ARD stigma, ARD exposure, self-perceived ARD risk, and demographic information.

Results: Linear regression revealed that higher anticipated ARD stigma accounted for 8.5% of variance in dementia-related anxiety. Hierarchical regression tested the primary hypothesis after controlling for demographic variables (Step 1: age, gender, years of education, relationship status, and general health) and ARD variables (Step 2: self-perceived ARD risk and ARD exposure). Step 3 included anticipated ARD stigma. The full model accounted for 47.3% of variance in dementia-related anxiety. Being female, reporting lower general health, higher self-perceived ARD risk, and greater anticipated ARD predicted greater dementia-related anxiety.

Conclusions: Given the cognitive focus of negative age-related stereotypes, it is perhaps not surprising that ARD, diseases associated with cognitive decline and later life, are stigmatized. Negative perceptions about ARD contribute to dementia-related anxiety, along with greater self-perceived ARD risk and poorer self-perceived physical health. Interventions aimed at reducing the stigma associated with aging and ARD may reduce dementia-related anxiety as well.

**THE NEUROTROPIC PARASITE TOXOPLASMA GONDII GENERATES A POPULATION OF MYELOID-LINEAGE CELLS THAT ARE EFFECTIVE AGAINST AMYLOID BETA DEPOSITION.** McGovern KE, Koshy AA. University of Arizona; Arizona Alzheimer's Consortium.

Background: Genetic and pathologic data suggest that amyloid beta (A $\beta$ ), produced by processing of the amyloid precursor protein, is a major initiator of Alzheimer's disease (AD). To gain new insights into A $\beta$  modulation, we sought to harness the power of the coevolution between the neurotropic parasite *Toxoplasma gondii* and the mammalian brain. Studies from our lab show that distinct, cyst-forming strains of *Toxoplasma* can manipulate the immune system of the host and generate varying populations of phagocytic myeloid cells. Here, we explored whether these cells can clear A $\beta$  in the context of infection despite historical evidence that suggests CNS inflammation contributes to the development of AD.

Methods: We infected an AD mouse model with genetically distinct, cyst forming strains of *Toxoplasma* (type II, or type III) and assessed plaque burden, immune cell infiltration, and the capacity for infiltrating phagocytes to take up A $\beta$ .

Results: We found that despite both type II and type III strains establishing a chronic CNS infection and inflammatory response, only type II infection was protective against A $\beta$  deposition. Both strains elicited increased numbers of CNS T cells and myeloid lineage cells, but infiltrating myeloid cells in the type II environment took up A $\beta$  most efficiently. These cells not only delay plaque deposition in young mice, but also reduce the size of plaques that are already established.

Conclusions: These data suggest that protective (type II) *Toxoplasma* strains generate a greater population of infiltrating monocytes that efficiently take up amyloid compared to nonprotective (type III) strains. Current work is focused on A $\beta$ 's interaction with the parasite, whether parasite burden is impacted by the level of A $\beta$  in the brain, and whether parasite effector proteins are sufficient for the creation of effector myeloid cell populations.

**APOE STATUS CORRELATES WITH THE ABILITY TO DETECT PHISHING EMAILS IN OLDER ADULTS.** McVeigh K, Hakim ZM, Wank AA, Ebner NC, Getz SJ, Levin BE, Wilson RC, Grilli MD. University of Arizona; University of Florida; University of Miami; Arizona Alzheimer's Consortium.

Background: Phishing emails constitute a major public health problem, linked to negative health outcomes due to fraud and exploitation. Because of their sheer volume, and because phishing emails are designed to deceive, purely technological solutions can only go so far, leaving human judgment as the last line of defense. However, because it is difficult to phish people in the lab, little is known about the cognitive and neural factors underlying phishing susceptibility and how these change with age. Recently, we developed an ecologically valid lab-based measure of phishing susceptibility. In the present study, we evaluated, for the first time, the sensitivity of this test to older age and another risk factor for Alzheimer's disease, namely apolipoprotein E (APOE).

Methods: In the Phishing Email Suspicion Test (PEST), participants rate a series of phishing and non-phishing emails according to their level of suspicion. Here, 78 cognitively unimpaired middle-aged to older adults (57 female, 21 male, mean age 68, range 47-83) performed 160 trials of PEST in the lab. In addition, participants completed standard neuropsychological assessment and were genotyped for APOE status.

Results: Initial findings suggest an interaction between APOE status and age such that the ability to detect phishing emails declines with older age in APOE4 carriers, but is approximately constant in middle-aged and older non-carriers.

Conclusions: This suggests that the presence of APOE4 may predict phishing susceptibility among cognitively unimpaired middle-aged and older adults. Older individuals at increased genetic risk for developing Alzheimer's disease dementia may be more vulnerable to phishing, and by extension, fraud and information or financial exploitation

**THE IMMUNOMETABOLIC CRISIS IN THE FEMALE AGING BRAIN: IMPLICATIONS IN ALZHEIMER'S DISEASE.** Mishra A, Shang Y, Wang Y, Bacon E, Yin F, Brinton RD. University of Arizona; University of Southern California; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease (AD) is characterized by a long latent prodromal stage with recent discoveries pointing to the perimenopausal transition in women as a "tipping point" in the development of the AD phenotype (Brinton et al., 2015). The hallmark chronic low-grade inflammation in both aging and menopause has been implicated as a unifying factor that bridges across these risk factors for AD (Mishra and Brinton, 2018). Yet, the endocrine state specific effect of female aging on neuroinflammation has not been characterized. In this study, we characterize the neuroinflammatory profile across chronological and endocrinological aging transitions with relevance to AD and neurodegenerative inflammatory mechanisms.

Methods: The perimenopausal rat model was used to study the effects of endocrinological and chronological aging distinctively. Unbiased transcriptomic analyses were conducted using bulk RNA Seq of the hippocampus. Further validation was done using immunohistochemistry and cell-specific assays. Clinical and translational validation was conducted using clinical datasets.

Results: Preliminary findings from the perimenopausal rat model, in which we can experimentally segregate the effects of chronological aging from endocrine aging, indicate that the inflammatory phenotype is extremely dynamic throughout the endocrine transition in menopause. Our results show that type I and type II interferon (IFN) response genes, recently implicated in age related neurodegeneration (Mathys et al., 2017) are upregulated in the hippocampus when the rats cycle irregularly. Co-incident with the upregulation of the IFN response genes in the hippocampus, was the overexpression of major histocompatibility complex (MHC) -II genes in white matter tracts – corpus callosum and fimbria. Reproductive irregularity also affected phagocytic response and redox status of microglial cells. Endocrine aging was associated with shifts in mitochondrial function in astrocytes and microglia. Estradiol regulation of the upregulation of interferon response genes was validated by ovariectomy and estradiol prevention paradigm. Clinical microarray data from the hippocampus was also analyzed to accomplish translational validity of the findings and, establish if the upregulation of MHC-II was preferentially observed in females.

Conclusions: The characterization of inflammation in the female aging brain and its role in transition to AD vulnerability has been, thus far, scarce. This pioneering study elucidates the dynamic immune decline in steroid hormones and brain glucose metabolism . Molecular characterization of the neuroinflammatory mechanisms during this neuro-endocrine transition state can inform therapeutic strategies to mitigate the risk of onset of Alzheimer's disease in women.

Acknowledgement: This work was supported by NIA P01AG026572 to RDB.

**DEVELOPING THERAPEUTICS TARGETING TDP-43 TO REDUCE AD PATHOLOGY.**  
Mollasalehi N, Francois-Moutal L, Scott DD, Mahoney B, Li X, Dong Y, Miranda V, Khanna R, Gokhale V, Barmada S, Clubb R, Wang W, Khanna M. University of Arizona; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

Background: Today 5.8 million people in the United States live with Alzheimer's disease (AD), including 1 in 10 of those 65 and over, estimates the Alzheimer's Association. It is the fifth leading cause of death in that age group. Despite growing investment by the NIH (\$2.4 billion in 2019) and pharma, Alzheimer's research has failed to deliver a cure, let alone a disease-slowing treatment. Dogmatic belief in the amyloid hypothesis, for example, has stifled emergence of new ideas. We advance a new approach: Allosterically modulating TDP-43 and effect on Tau.

TAR-DNA-binding protein-43 (TDP-43) is a nuclear protein implicated in transcriptional repression and splicing. The protein was reported to be present in inclusions in the neurons and/or glial cells of a range of neurodegenerative diseases, including AD. In diseased neurons with TDP-43-positive inclusions, TDP-43 is improperly processed and/or transported. Depletion of cellular TDP-43 molecules, as a result of their being trapped in inclusions, leads to the loss of neuronal activities regulated by TDP-43. The inclusions also cause impairment of mitochondrial function and neuronal cell death. TDP-43 pathology is comorbid with tau, a major microtubule associated protein. TDP-43 has also been linked to Tau splicing and mRNA stability.

Methods: We target in silico the N-terminal domain of TDP-43 in an attempt to overcome the pathologic synergy of these proteins.

Results: We will present unpublished data of the interaction of TDP-43 with Tau and define how an allosteric molecule could influence their interactions.

Conclusions: Our preliminary data shows a nanomolar interaction between Tau and TDP-43. Additionally, through N-terminal targeting of TDP-43, we discovered a molecule that allosterically modulates RNA-binding.

**THE ROLE OF ONE-CARBON METABOLISM ON STROKE OUTCOME IN AN AGED MOUSE MODEL.** Mosnier H, Kelly E, Lawrence K, Cruickshank S, Stacey S, McCall A, Dhatt S, Arning E, Bottiglieri T, Smith PD, Jadavji NM. Carleton University; Baylor Scott & White Research Institute; Midwestern University; Arizona Alzheimer's Consortium.

**Background:** Nutrition is a modifiable risk factor for stroke, which is one of the leading causes of death and disability world-wide. In humans deficiencies in one-carbon metabolism, including the methyltetrahydrofolate reductase (MTHFR) polymorphism, have been linked to increased risk of stroke. The Mthfr+/- mice mouse model mimics the phenotype of the MTHFR677C -- >T polymorphism. In our work using in vitro and in vivo models of ischemic stroke we have observed decreased recovery after stroke through reduced neuronal and astrocyte viability and increased apoptosis in MTHFR-deficient mice. In addition, we have previously shown dietary supplementation of one-carbon metabolites increases neuroplasticity and reduced oxidative stress after ischemic stroke. Using the MTHFR-deficient mouse model, the aim of this study was to investigate the impact of dietary supplementation with one-carbon metabolites on stroke outcome.

**Methods:** Male Mthfr+/- and wildtype littermate control mice were aged to 1.5-year-old and were placed on control diet (CD) 4-weeks prior to sensorimotor cortex damage using photothrombosis (PT), a model for ischemic stroke. Post-operatively, one group of Mthfr+/- and wildtype littermate mice were fed a supplemented diet (SD) containing 5-methylTHF, vitamin B12, and choline. Four weeks after PT damage and SD motor function was assessed and brain tissue was processed to assess lesion volume and investigate biochemical and molecular changes.

**Results:** Mthfr+/- mice fed a SD after PT did not have an impaired neuroscore compared to CD Mthfr+/- mice. When compared to CD, SD Mthfr+/- mice were able to stay on the accelerating rotarod longer and travelled further, they also used their impaired forepaw more. Total homocysteine levels in plasma and lesion volume were reduced in SD Mthfr+/+ and Mthfr+/- mice. In the brain, within the damage site, there were reduced levels of apoptotic cell death and an increased neuroprotective cellular response in SD treated Mthfr+/- mice.

**Conclusions:** This study reveals a critical role for one-carbon supplementation in supporting improvement of function after ischemic stroke. Our data suggests that in stroke affected patients, nutritional supplementation maybe an important component to post-operative care, in addition to pharmacological and other rehabilitation therapies.

**PSEUDOTIME ANALYSIS IDENTIFIES OLIGODENDROCYTE GENES ASSOCIATED WITH CLINICAL AND NEUROPATHOLOGICAL PROGRESSION TO ALZHEIMER'S DISEASE.** Piras IS, Naymik MA, Huentelman MJ. Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

Background: Pseudotime algorithms allow extraction of latent temporal information from cross-sectional studies related to biological processes that should be ideally studied using longitudinal frameworks. However, longitudinal studies are often limited by logistical and financial barriers. This is especially true with brain diseases as tissue availability is limited to a single timepoint represented by the autopsy. The "PhenoPath" method allows the extraction of unidimensional pseudotime trajectories modulated by a factor of interest. We applied this algorithm to a public Alzheimer's Disease (AD) RNA-sequencing dataset from post-mortem brains including non-demented donors and AD patients with different neuropathological states of the disease.

Methods: RNA-sequencing and clinical data from the Mount Sinai study were downloaded from (<https://www.synapse.org/#syn7391833>). We focused on the parahippocampal gyrus (BA36), a region that shows the largest RNA expression dysregulation. Then, we selected the oligodendrocyte specific genes using our recently published bulk deconvolution method. Genomic trajectories were inferred by incorporating Clinical Dementia Rating (CDR) and Braak Scores separately, using the following parameters as implemented in the phenopath function in the homonym R package: `elbo_tol = 1e-5`, `Z_init = "random"`, `thin = 10`, and `maxiter = 10E4`. Associations of total expression profiling with pseudotime and covariates were assessed by Principal Component Analysis (PCA). The relationship of gene expression with neuropathological variables and pseudotime was assessed by linear regression with an interaction model.

Results: A total of 355 oligodendrocytes specific genes were selected from the total dataset. We found a significant correlation of the first two Principal Components (PCs) with both CDR and Braak Score, as well as with pseudotime. A total of 36 genes were associated with both CDR and pseudotime, whereas 21 genes were associated with both Braak Stage and pseudotime. Interestingly, 13 genes were overlapping between the CDR and Braak Stage analyses. Among the genes associated, we found NKX6-2, SLC45A3, ENPP2, ASPA and PLP1, genes involved in oligodendrocyte differentiation and axon ensheathment.

Conclusions: Our preliminary results highlight oligodendrocyte genes showing correlation with neuropathological and clinical variables and disease progression as assessed by latent temporal trajectories. Several lines of evidence suggest a role for oligodendrocyte dysfunction in AD prior to the appearance of amyloid and tau pathology. Further studies are needed to evaluate the specific role of these genes and their potential contribution to the earliest stages of the disease. Additionally, this work suggests that oligodendrocyte supportive therapeutic approaches should be explored as potential treatments for AD.

**A NOVEL LONGITUDINAL APPROACH IN INDIVIDUAL SUBJECTS TO INVESTIGATE THE MOVEMENT OF TAU OVER TIME USING GRAPH THEORY IN CLINICAL AND PRECLINICAL STAGES OF ALZHEIMER'S DISEASE.** Protas HD, Ghisays V, Chen Y, Goradia DD, Luo J, Malek-Ahmadi MH, Lee W, Devadas V, Bauer III R, Reiman EM, Chen K, Su Y, Banner Alzheimer's Institute; University of Arizona; Translational Genomics Research Institute; Arizona State University; Arizona Alzheimer's Consortium.

Background: We previously introduced an index characterizing inter-regional tau deposition variability based on graph theory. Extending our method to additionally characterize the longitudinal tau spread for individual subjects based on directed graph theory, we examined the difference between longitudinal network measures and the regional tau deposit measures over time with respect to 1) differentiation of patients with AD, MCI, and cognitively unimpaired(CU) subjects, 2) correlation with memory measures in A $\beta$ + CU, and 3) sample size needed for a prevention trial for A $\beta$ +/- CU.

Methods: The longitudinal tau PET data is from 13 AD, 42 MCI, and 72 CU in ADNI . Using cerebellar grey reference region, we calculated the difference over two time points (1.1 years  $\pm$  0.2 (0.6-1.6)) for the entorhinal, inferior temporal and metaROI SUVR, and the difference for indices, in-strength and out-strength each based on the directed tau longitudinal network using predefined nodes(inferior temporal and limbic). We examined regional/network based indices in term of the group differences, their correlation with AVLT-LTM change. In addition, we also calculated sample size for a prevention clinical trial for A $\beta$ +/- subjects separately with 80% power and 25% treatment effect.

Results: The longitudinal tau network measures(instrength and outstrength) are significantly different between the three groups ( $p < 0.004$ ) but not the SUVR change at entorhinal( $p = 0.62$ ), inferior temporal( $p = 0.36$ ) or tau meta ROI rates( $p = 0.21$ ). In CU A $\beta$ + subjects, limbic outstrength significantly correlated to AVLT-LTM rate( $r_s = -0.52$ ,  $p = 0.013$ ). The most significant regional value(tau metaROI) rate is not significantly correlated with AVLT LTM rate( $r_s = -0.25$ ,  $p = 0.26$ ). For a clinical trial, the Inferior temporal in-out strength(net influx) requires a sample size of 584 subjects in CU A $\beta$ + group and a sample size of 26448 subjects in CU A $\beta$ - Entorhinal tau SUVR, inferior temporal and tau metaROI SUVR needs a sample size for CU A $\beta$ + is 1623, 2450, 2629 respectively. For CU A $\beta$ - subjects, the sample size is 53537 for entorhinal SUVR, 50605 for inferior temporal SUVR and 916184 for tau metaROI SUVR .

Conclusions: The network based longitudinal indices provide better power for differentiating patients at different stages, for examining tau's effects on memory and for detecting a treatment effects in prevention trial.

**APOE4 DISRUPTS LIPID METABOLISM ACROSS NEURON AND ASTROCYTE.** Qi G, Mi Y, Shi X, Gu H, Brinton RD, Yin F. University of Arizona; Arizona State University; Arizona Alzheimer's Consortium.

Background: The  $\epsilon 4$  variant of apolipoprotein E (ApoE4) is the greatest genetic risk factor for late-onset Alzheimer's disease (AD). ApoE4 is known to not only impair amyloid- $\beta$  (A $\beta$ ) clearance and promote its aggregation, but also perturb lipid homeostasis and energy metabolism in brain. However, the mechanistic role of ApoE4 in modulating the bioenergetic properties of lipid species in different brain cell types is unknown. Here we describe an isoform-specific- and cell-type-specific role of ApoE in regulating lipid accumulation and degradation across neuron and astrocyte, in connection with its diverse impact on neuronal and astrocytic mitochondrial function.

Methods: To determine the mechanism by which ApoE isoforms differentially regulate lipid metabolism across brain cell types, fatty acid (FA) metabolism, lipid droplet (LD) formation and mitochondrial function were determined in primary neurons and astrocytes isolated from forebrains of humanized ApoE3 and ApoE4 knockin mice.

Results: We observed substantial accumulation of neutral lipids in LDs in both neurons and astrocytes of either ApoE3 or ApoE4 genotype. Intriguingly, when compared to ApoE3 neurons, ApoE4 neurons sequestered less lipids in LDs, which was coupled with higher levels of free FAs. Our results further revealed that elevated FA levels contributed to mitochondrial dysfunction seen in ApoE4 neurons. In contrast to neurons, ApoE4 led to markedly higher lipid accumulation in astrocyte LDs relative to ApoE3, which was ascribed to the decreased capacity of ApoE4 astrocytes in metabolizing FAs via mitochondrial  $\beta$ -oxidation.

Conclusions: These data provide new insights into the mechanism of brain lipid metabolism across neuron and astrocyte, and support a vital role of lipotoxicity in the etiology of AD. Further, our findings reveal the mechanistic basis of ApoE4 in disrupting FA metabolism in brain that may underlie the exaggerated lipid dyshomeostasis and bioenergetic decline, and increased AD risk for ApoE4 carriers.

## **RELATION OF DAILY ACTIVITY PATTERNS TO CORTICAL GRAY MATTER MAPS IN THE HEALTHY OLDEST OLD: FINDINGS FROM THE MCKNIGHT BRAIN AGING REGISTRY.**

Raichlen DA, Bharadwaj PK, Franchetti MK, Sims S, Rezaei RF, Merritt S, Jessup CJ, Porges ES, Geldmacher D, Hishaw GA, Alperin N, Trouard TP, Wadley VG, Levin BE, Woods AJ, Rundek T, Visscher K, Cohen RA, Alexander GE. University of Arizona; University of Alabama at Birmingham School of Medicine; University of Miami Miller School of Medicine; University of Florida; William L. McKnight Brain Institute; Arizona Alzheimer's Consortium.

Background: Engaging in increasing levels of physical daily activity (PA), while having good sleep quality may help in maintaining cognitive and brain health during aging. Wrist-worn accelerometers provide a way to measure engagement in different aspects of daily activity, including levels of moderate to vigorous physical activity (MVPA), fractal patterns of consistent PA (FPA), as well as movement during sleep, reflecting sleep efficiency (SE). How these different measures of activity relate to brain health in oldest old adults has yet to be investigated. We sought to determine whether having high levels of MVPA, FPA, and SE are associated with greater cortical gray matter (GM) in a cohort of oldest-old adults from the McKnight Brain Aging Registry.

Methods: For this analysis, 64 community-dwelling, cognitively unimpaired older adults, ages 85 to 99 were included [mean±sd age = 87.9±3.3; M/F = 31/33; mean±sd Mini-Mental State Exam = 28.4±1.3]. Volumetric T1-weighted 3T MRI scans were acquired across the McKnight Brain Institutes at the University of Arizona, University of Alabama at Birmingham, University of Miami, and University of Florida - Gainesville. The MRI scans were processed using Freesurfer (v6.0) and total intracranial volume (TIV) was computed using SPM12 to adjust vertex-wise volume maps for head-size. Measures of MVPA, FPA, and SE were acquired with Actigraph accelerometers worn on the non-dominant wrist for up to seven consecutive days. MVPA, FPA, and SE were defined with standard algorithms using the GGIR package (v1.6.0) in R (v3.4.4). Analyses tested the relation of MVPA, FPA, and SE to cortical maps of thickness, area, and volume using extent thresholds to maintain an overall  $p < 0.05$  false positive rate (Greve & Fischl, 2018).

Results: Results showed that, after adjusting for TIV, higher levels of MVPA were significantly associated with increased volumes in the vicinity of left lateral temporal and medial frontal regions. Greater MVPA was also significantly associated with greater cortical thickness in parieto-occipital regions; greater FPA was associated with greater thickness in precentral and inferior parietal regions; and greater SE was associated with increased cortical area in inferior parietal regions. The regions of cortical volume, area, and thickness were also significantly associated with better cognitive performance.

Conclusions: Among cognitively unimpaired oldest old adults, engaging in more MVPA and FPA, and having better SE are each associated with enhanced gray matter, involving brain regions that are related to better cognitive functions. Together these results support the benefits of PA and sleep quality for brain health in the context of successful cognitive aging.

## **REMOTE ISCHEMIC CONDITIONING ACUTELY ATTENUATES PERIPHERAL INFLAMMATION AND MICROGLIAL ACTIVATION AFTER DIFFUSE BRAIN INJURY WITH LONG-TERM IMPACT ON BEHAVIOR AND INFLAMMATION IN BOTH SEXES OF MICE.**

Saber M, Hur Y, Christie I, Giordano KR, Young C, Tallent BR, Rowe RK, Lifshitz J. University of Arizona College of Medicine-Phoenix; Barrow Neurological Institute at Phoenix Children's Hospital; Phoenix VA Healthcare System; Arizona Alzheimer's Consortium.

**Background:** Remote ischemic conditioning (RIC) is the intermittent restriction of blood flow to a limb or non-vital organ. This therapeutic strategy protects major organs from ischemia-reperfusion injury, reduces cognitive impairments in vascular dementia models and traumatic brain injury (TBI), and halts the increase of acute biomarkers after severe TBI, and can reduce mortality in sepsis models. Though the mechanism of RIC is unknown, RIC may modulate the inflammatory response. We hypothesized that post-injury RIC would reduce the population of peripheral monocytes in the diffuse-injured brain acutely, have sustained therapeutic effect on cognitive performance and anxiety, and protect against a secondary inflammatory challenge.

**Methods:** Diffuse brain injury by midline fluid percussion or sham injury was performed on adult C57BL/6J mice of both sexes. After 1-hour, mice received 4×5-minute sessions of RIC (tourniquet on thigh) with a 5-minute reperfusion between each session or anesthesia control. Blood, spleen, and brains were collected 3 days post-injury (DPI; n=54). Brains and spleens were analyzed using immunohistochemistry, and plasma was used to analyze peripheral cytokine expression. Further, at 3 and 7DPI, blood, spleen, bone marrow, and brains were analyzed using flow cytometry to quantify monocyte, neutrophil, and macrophage populations (n=76). A separate cohort (n=104) was used to test the therapeutic efficacy of RIC on cognitive performance and anxiety over 90DPI. We hypothesized that previous RIC treatment may protect against a secondary inflammatory challenge. Therefore, mice were administered lipopolysaccharide (10 mg/kg LPS, i.p.) at 100DPI. Sickness behavior, peripheral inflammation, and neuroinflammation were measured.

**Results:** RIC-treated TBI mice had significantly fewer peripheral monocyte populations in the blood and spleen compared with non-RIC treated TBI mice at 3DPI, though there were no changes in peripheral cytokine expression. These findings extended to the brain; RIC reduced peripheral macrophage populations in RIC-treated TBI mice compared with non-RIC treated TBI mice. Furthermore, RIC treatment reduced TBI-induced microglial activation in the dentate gyrus to sham levels compared with TBI controls. Lastly, though RIC-treated TBI and non-RIC TBI had similar motor deficits as measured through rotarod testing, RIC treated TBI mice had a higher differential index in novel object recognition (NOR) testing compared to non-RIC treated TBI mice and non-RIC TBI mice showed a deficit in NOR compared to sham mice.

**Conclusions:** RIC remains a practical, personalized therapy for TBI, in part by reducing neuroinflammation and peripheral inflammation.

These works were supported by R21-NS096515 and Arizona Alzheimer's Consortium.

**CORRELATION BETWEEN PARTICIPANT DEMOGRAPHICS AND WILLINGNESS TO PARTICIPATE IN ALZHEIMER'S PREVENTION CLINICAL TRIALS: RESULTS FROM GENEMATCH RECRUITMENT INTO THE API GENERATION PROGRAM.** Salata H, Gordon D, Stowell C, Walsh T, High N, Kettenhoven C, Reiman EM, Tariot PN, Langbaum JB. Banner Alzheimer's Institute; Provo, Washington, DC; Arizona Alzheimer's Consortium.

Background: The API Generation Program consists of two trials: Generation Studies 1 and 2. Generation Study 1 enrolled cognitively unimpaired (CU) APOE4 homozygotes (HMs) ages 60-75; Generation Study 2 enrolled CU APOE4 carriers (HMs and heterozygotes (HTs), HTs must also have elevated brain amyloid). We developed GeneMatch to support recruitment into the Generation Program as well as other studies.

Methods: GeneMatch is a US-based, trial-independent recruitment registry, performing APOE genotyping in US-based CU individuals aged 55-75 years to enrich referrals to prevention studies. Buccal swabs are sent to participants' homes or distributed at partner sites after online enrollment. GeneMatch does not disclose APOE results to participants directly, however, APOE results are used to match and invite participants to enrolling studies in a manner that does not inadvertently disclose results. We analyzed data from GeneMatch participants invited to the Generation Program to identify whether age, sex, self-reported family history of AD or other dementia, distance from participant's home to the closest study site, or time between GeneMatch enrollment and receiving a Generation Program study invitation were correlated with invitation acceptance.

Results: To date, >90,000 participants have joined GeneMatch, mean age 65 years (SD 5.4), 69% women, >30% APOE4 carriers. Recruitment in the Generation Program and treatment with umibecestat was terminated in July 2019 after an early signal of mild worsening in some measures of cognitive function with umibecestat. At that time, 16,521 GeneMatch participants had been invited to the Generation Program, 5,461 of whom had accepted their study invitations. Weak correlations to study acceptance were found. Time was negatively correlated (-0.17) with invitation acceptance, as was distance (-0.07). Family history was positively correlated with invitation acceptance (0.11). Age and sex were not correlated with invitation acceptance after accounting for time in GeneMatch to invitation.

Conclusions: GeneMatch successfully helped with recruitment in the US for the Generation Program. Moving forward, recruitment into GeneMatch should mirror the geographic footprint of study sites. In addition, it will be important to pace enrollment into GeneMatch so that participants do not have to wait long periods of time before a nearby site is available for trial enrollment.

**INHIBITING RIPK3-MLKL PROTEIN-PROTEIN INTERACTIONS IN NECROPTOSIS ACTIVATION.** Sanchez J, Gokhale V, Khanna M. University of Arizona; Arizona Alzheimer's Consortium.

Background: Necroptosis, a form of programmed necrosis, is activated in several diseases ranging from pulmonary, hepatic, cardiovascular, and renal diseases. More recently, necroptosis has been observed to contribute to neuronal loss in several neurodegenerative diseases such as Alzheimer's disease, Parkinson's disease, and Amyotrophic lateral sclerosis. Activation of Necroptosis occurs through three key proteins, RIPK1, RIPK3, and MLKL, responding to inflammation factors. RIPK1 and RIPK3 are kinases that are activated and subsequently bind and activate MLKL, which is the effector protein of cell death through direct or indirect methods by breaking down the cellular membrane. In order to prevent neuronal cell death, a binding pocket within RIPK3 was targeted to prevent MLKL binding, as mutations in MLKL (F373) known to bind the RIPK3 pocket prevent complex formation. To identify possible compounds that may bind the RIPK3 pocket and thereby prevent MLKL from binding, in silico docking was performed using the Schrodinger suite of programs.

Methods: Necroptosis was induced in HT-29 cell lines by addition of 25 ng/mL TNF- $\alpha$ , 20  $\mu$ M Z-VAD-fmk, and 0.1  $\mu$ M SM-164 to the media. Additionally, 100  $\mu$ M of putative compounds identified through the Schrodinger suite of programs were also added to the media. After 7 hrs, HT-29 cells were stained with Annexin V and Propidium Iodide (PI) and analyzed by FACS to identify necroptosis positive cells. Compounds found to inhibit necroptosis in HT29 cells were utilized in a Cellular Thermal Shift Assay (CETSA) to identify the Tagg of the compounds, thus indicating compounds are binding and stabilizing RIPK3 in a cellular context as a thermal gradient is applied. Initial compounds identified to inhibit necroptosis and bind the RIPK3 were analyzed in order to identify commercially available derivatives in order to identify a structure activity relationship. Derivative compounds were analyzed similarly as previously indicated in FACS analysis. The originally identified necroptosis inhibitors alongside the top performing compound derivatives, as determined by FACS, were used in a cell viability assay utilizing alamarblue to obtain an EC50 in necroptosis induced HT29 cells.

Results: In silico docking identified 13 putative compounds that could bind to the RIPK3 pocket and possibly disrupt RIPK3 and MLKL complex formation. Flow cytometry analysis of necroptosis induced HT29 cells resulted confirmed only 2 of the 13 compounds could inhibit necroptosis. These two compounds were also found to bind to RIPK3 in cellular context as a shift the thermal aggregation temperature was observed in a CETSA experiment. Derivates of the two compounds were analyzed by FACS and most potent inhibitor from each of the two compounds were used in a cell viability experiment to obtain EC50 values. The EC50s indicated that improvements have been discovered at least in part for one of the compounds while ongoing screening is being performed to identify more potent inhibitors.

Conclusions: We have identified two structurally distinct compound entities that are able to bind RIPK3 and inhibit necroptosis. Ongoing screening have obtained improvements for these original compounds in order to inhibit necroptosis. With further optimizations, these small molecules may provide a novel means of inhibiting necroptosis through disruption of protein protein interactions not only in several neurodegenerative diseases but also in other necroptosis associated diseases.

**AMYLOID- $\beta$  PLAQUES IN THE OLFATORY BULB IN RELATION TO THE PATHOLOGIC PROGRESSION OF THAL AMYLOID STAGE IN ALZHEIMER'S DISEASE.** Serrano GE, Russell A, Vargas D, Intorcica A, Walker J, Glass M, Arce R, Oliver J, Papa J, Nelson C, Sue LI, Beach TG. Banner Sun Health Research Institute; Arizona Alzheimer's Consortium.

Background: Thal amyloid stage is correlated with cognitive function and is a measure of the progression of Alzheimer's disease (AD) within the brain. Higher Thal stages ( $> 3$ ) are strongly predictive of the presence of diagnostic levels of Alzheimer's disease neuropathological change (ADNC). Previous work has shown that alpha-synuclein, tau and amyloid- $\beta$  ( $a\beta$ ) can be detected in the olfactory bulb and tract (OBT) of elderly individuals. However, to our knowledge no study has evaluated OBT  $a\beta$  pathology with respect to its possible value for predicting Thal amyloid stage and dementia due to ADNC.

Methods: Autopsied subjects with available Thal amyloid staging were selected by a database search of the Arizona Study of Aging and Neurodegenerative Disorders (AZSAND) and Brain and Body Donation Program (BBDP). We analyzed neuropathologically-confirmed cases of ADD (N = 35), and no ADD (N= 30). ADD was defined as the presence of dementia with Intermediate or High ADNC (moderate or frequent CERAD cortical neuritic plaque density and Braak neurofibrillary stage III-VI). The OBT were stained for  $A\beta$  using immunoperoxidase methods (6E10 antibody); results were graded using CERAD templates. Demographic data were compared with t-tests and Fisher exact tests. Diagnostic accuracy was estimated by calculating sensitivity and specificity.

Results: There were no differences in age at death or gender ratios between ADD and non-ADD. The presence of OBT  $A\beta$  deposits was 82.6% sensitive and 75.5% specific for predicting Thal  $A\beta$  stage  $> 3$ , with a positive predictive value of 63.3% and a negative predictive value of 87.9%.

Conclusions: These results suggest that olfactory bulb biopsies may be useful for detecting diagnostic levels of multiple different brain pathologies, including Thal amyloid stage.

**WHOLE SOMA DISSOCIATED SUSPENSION ANALYSIS IN HUMAN BRAIN NEURODEGENERATIVE DISEASE: A PILOT STUDY.** Serrano GE, Walker J, Intorcchia A, Glass M, Arce R, Piras I, Talboom J, Oliver J, Nelson C, Papa J, Cutler B, Sue LI, Lue L, Huentelman MJ, Beach TG. Banner Sun Health Research Institute; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

Background: Biochemical analysis of human neurodegenerative brain tissue is typically done by homogenizing whole pieces of brain and separately characterizing the proteins, RNA, DNA and other macromolecules within. While this has been sufficient to identify large changes, there is very little ability to identify small changes, or changes in small subsets of cells. To effectively investigate the biochemistry of neurodegenerative disease in the brain, with its thousands of different cell types, we must first separate the cells, and study them as phenotypically-defined populations and even as individuals.

Methods: In this project we developed a new method for the generation of whole-soma-dissociated-suspensions (WSDS) in fresh human brain that could be shared with scientists as a resource to study human single cells or cell types populations. Characterization of WSDS was done in paraffin-embedded sections stained with H&E, by immunophenotyping with antibodies specific and by fluorescence-activated cell sorting (FACS) utilizing the same antibodies. Additionally, we compared extracted RNA from WSDS with RNA from adjacent intact cortical tissue, using RT-qPCR for cell-type-specific RNA for the same markers as well as whole transcriptome sequencing.

Results: Each examined dissociated cell suspension always had a diverse population, typically including approximately 40% neurons, 25% astrocytes, 21% microglia, 5% oligodendrocytes and 4% endothelial cells. RNA integrity number (RIN) for unfixed WSDS incubated for 4hrs in enzyme ranged from 2 to 8, with a mean RIN of 6.2 and 2.1 standard deviation. The yield of RNA ranged from 4-350ng/million cells, with a mean yield of 55 ng/million cells. RNA extraction from WSDS and WTH were done on twelve random selected cases. RIN mean for those 12 WSDS was 6.3 +/-2.0, while WTH had a RIN mean of 6.5 +/-2.0. q-PCR results suggest that neuronal NEU-N and astrocyte GFAP RNA expression was not different between WSDS and WTH, while RNA expression of the well know microglia protein IBA1 was upregulated in WSDS. More than 11,626 gene transcripts were successfully sequenced and classified either as being mainly expressed in neurons, astrocytes, microglia, oligodendrocytes, endothelial cells, or mixed (in two or more cell types).

Conclusions: Our results demonstrate that we are currently capable of producing WSDS with full representation of different brain cell types together with RNA quality suitable for use in biochemical analysis.

**EVIDENCE IN SUPPORT OF CHROMOSOMAL SEX INFLUENCING PLASMA BASED METABOLOME VS APOE GENOTYPE INFLUENCING BRAIN METABOLOME PROFILE IN HUMANIZED APOE MALE AND FEMALE MICE.** Shang Y, Mishra A, Wang T, Wang Y, Desai M, Chen S, Mao Z, Do L, Bernstein AS, Trouard TP, Brinton RD. University of Arizona; University of Southern California; Arizona Alzheimer's Consortium.

Background: Late onset Alzheimer's disease (LOAD) is a progressive neurodegenerative disease with four well-established risk factors: age, APOE4 genotype, female chromosomal sex, and maternal history of AD. Each risk factor impacts multiple systems, making LOAD a complex systems biology challenge.

Methods: To investigate interactions between LOAD risk factors, we performed multiple scale analyses, including metabolomics, transcriptomics, brain magnetic resonance imaging (MRI), and beta-amyloid assessment, in 16 months old male and female mice with humanized human APOE3 (hAPOE3) or APOE4 (hAPOE4) genes.

Results: Metabolomic analyses indicated a sex difference in plasma profile whereas APOE genotype determined brain metabolic profile. Consistent with the brain metabolome, gene and pathway-based RNA-Seq analyses of the hippocampus indicated increased expression of fatty acid/lipid metabolism related genes and pathways in both hAPOE4 males and females. Further, female transcription of fatty acid and amino acids pathways were significantly different from males. MRI based imaging analyses indicated that in multiple white matter tracts, hAPOE4 males and females exhibited lower FA than their hAPOE3 counterparts, suggesting a lower level of white matter integrity in hAPOE4 mice. Consistent with the brain metabolomic and transcriptomic profile of hAPOE4 carriers, beta-amyloid generation was detectable in 16-month-old male and female brains.

Conclusions: These data provide therapeutic targets based on chromosomal sex and APOE genotype. Collectively, these data provide a framework for developing precision medicine interventions during the prodromal phase of LOAD, when the potential to reverse, prevent and delay LOAD progression is greatest.

**A NEW METHOD TO QUANTIFY THE DYNAMICS OF THOUGHT AND EMOTION REVEALS EFFECTS OF AGE AND DEPRESSIVE SYMPTOMS.** Spillman A, Griffith C, Andrews E, Rafaelli Q, Strojek D, Tran A, Hernandez A, Majeed Z, Lehner A, Wilcox R, Matijevic S, Robertson A, Mehl M, O'Connor MF, Ryan L, Grilli M, Andrews-Hanna JR. University of Arizona; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease (AD) is associated with major alterations in thought and emotion, including impairments in episodic memory retrieval and difficulties regulating emotion. Typically, these processes are quantified using experimentally-directed paradigms that constrain the focus of thought over multiple discrete trials. Consequently, very little is known about spontaneous alterations in thought and emotion, nor about the ways in which thoughts and feelings arise and unfold naturally over time (that is, their dynamic properties). In order to better understand the spontaneous, dynamic nature of thought and its relevance for AD risk, we adapted a new "Think Aloud" method and applied the method to two AD risk factors: age and depressive symptoms.

Methods: We adapted a "Think Aloud Task" for use in an MRI machine in the service of a broader goal to understand the neural underpinnings of thinking styles (MRI analyses not discussed here). The task requires that younger and older adults verbalize their unprompted and uncensored thoughts for 7 minutes while they lie in an fMRI scanner. Audio recordings of participants' responses were then rated dynamically over the 7 minutes by unbiased coders for different cognitive and emotional constructs such as thought specificity, attention orientation (internal stimuli vs external stimuli), positive valence, and negative valence. Participants also filled out questionnaires related to depressive symptoms, which allowed us to determine if there were any associations between the questionnaire scores and the Think Aloud ratings.

Results: Preliminary analyses assessed the relationships between dynamic ratings of younger and older adults with and without depressive symptoms, using the 4 separate rating constructs. Older adults' thoughts were significantly more positive than younger adults, supporting an age-related positivity effect. Independent of age, increased depressive symptoms was linked to less positive, more negative and less specific thoughts.

Conclusions: Although these findings are preliminary, they suggest that a stream of consciousness Think Aloud task may be an effective means with which to assess the dynamics of spontaneous thought in relation to AD risk and possibly disease progression. Further analysis may offer additional insights into how AD risk factors are reflected in everyday thoughts, thus facilitating the development of new therapeutic interventions.

**TMS REVEALS DIMINISHED HOMEOSTATIC METAPLASTICITY IN NON-DEMENTED OLDER ADULTS WITH COGNITIVE IMPAIRMENT.** Sundman MH, Lim K, Ton That V, Mizell JM, Ugonna C, Rodriguez R, Chen NK, Fuglevand AJ, Wilson RC, Fellous JM, Rapcsak S, Chou YH. University of Arizona; Arizona Alzheimer's Consortium.

Background: Homeostatic metaplasticity is a neuroprotective physiological feature that acts as a counterbalance to Hebbian forms of plasticity to prevent network destabilization and hyperexcitability. Recent animal models highlight dysfunctional homeostatic metaplasticity in the pathogenesis of Alzheimer's Disease. Herein, we sought to investigate this neurophysiological feature and its association with cognitive status in a population of non-demented older adults.

Methods: 40 healthy older adults enrolled in this study (age range: 65-74, 19 females). Cognitive function was assessed with the NACC UDS 3.0 battery. To assess neural plasticity, we interleaved single-pulse transcranial magnetic stimulation (TMS) measures with a bout of excitatory intermittent theta-burst stimulation (iTBS). Single-pulse measures were sampled at multiple intensities to generate an input/output curve. There were two experimental paradigms: one with voluntary muscle contraction to deliberately introduce homeostatic interference, and one without to serve as a control condition. We compared neuroplastic responses across these experimental paradigms, and across cohorts grouped by cognitive status.

Results: When examining iTBS responses, significant interaction effects were observed across cognitive status and experimental paradigm at multiple levels of the input/output curve (e.g., @TMS intensity of 140% motor threshold,  $F(1,36) = 8.2$ ,  $p < 0.01$ ). When considering motor evoked potentials across all TMS intensities sampled, findings differed in across the two paradigms. In the active paradigm, a 3-way ANOVA revealed a significant interaction effect across cognitive status, iTBS, and TMS intensity ( $F(15,270) = 3.1$ ,  $p < 0.001$ ). The interaction effects consistently indicate that homeostatic interference blunted the otherwise excitatory effect of iTBS in the cognitively normal cohort, but not in the cognitively impaired cohort. No significant effects were observed in the rest paradigm.

Conclusions: We report that homeostatic metaplasticity is diminished in our cohort of cognitively impaired older adults, but this neuroprotective feature remains intact in cognitively normal participants. This finding was observed in the presence of no significant difference in Hebbian-like plasticity across cognitive status in this population of non-demented older adults. This novel finding suggests future studies should expand their scope beyond just the Hebbian forms of plasticity that are traditionally assessed to also probe homeostatic metaplasticity when investigating neural plasticity in older adults. This may be a useful new paradigm to detect early physiological changes associated with poor cognitive aging.

**FIRST DEGREE FAMILY HISTORY OF ALZHEIMER'S DISEASE INFLUENCES SIMPLE VISUAL REACTION TIME PERFORMANCE AND IS MODIFIED BY HEALTH FACTORS.**

Talboom JS, Schmidt AM, De Both MD, Håberg AK, Naymik MA, Schrauwen I, Jepsen WM, Lewis CR, Moore B, Karaniuk K, Metz D, Raine M, Fritz MA, Myers AJ, Hay M, Barnes CA, Glisky E, Ryan L, Huentelman MJ. Translational Genomics Research Institute; Norwegian University of Science and Technology; University of Miami; University of Arizona; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease (AD), the leading cause of dementia, is a progressive neurodegenerative disease that first presents itself clinically as deficits in cognition, including learning and memory. Currently, more than 5 million Americans are afflicted, costing over 315 million dollars. To this day, despite numerous clinical trials, there are no disease-modifying treatments, leaving risk reduction as the only currently viable means to slow or stop AD from turning into a chronic public health emergency. Developing novel risk reduction approaches requires finding and understanding the complex environmental and genetic interactions leading to AD. To that end, we developed MindCrowd ([www.MindCrowd.org](http://www.MindCrowd.org)), a fun and easy-to-use web-based study. Consenting participants take one test of simple visual reaction time (svRT), one test of paired associates learning, and answer 23 demographic, health, and lifestyle questions. To date, over 71,424 individuals, spread across the globe, have participated, with ages ranging from 18-100.

Methods: After this data was filtered (e.g., removing outliers & participants with brain diseases), our final cohort consisted of 69,774 participants. Outliers were identified using a standard method widely used across disciplines. Specifically, trials or participants with svRT values 1.5 times the interquartile range greater or less than from the third or first quartile respectively were removed. Outlying trials were removed within each subject, and outlying participants were excluded based on the median svRT values of participants of the same age. The median svRT (ms), recalculated after outlier removal, served as our "criterion" (dependent variable) in multiple regression analysis (general linear model). All 23 demographic, health, and lifestyle questions were entered into our multiple regression analysis. A subset of these questions served as our "predictors" (independent variables), and the rest were used as "control variables" or covariates.

Results: We found that sex was associated with svRT, and FH was associated with slower svRT beginning around the fourth decade of life. Further, there was an inverse association between Educational Attainment and svRT. Lastly, akin to our prior published finding in paired associates learning, we discovered that diabetes modified the association of FH on svRT. Diabetes, in conjunction with FH, appears to have a synergistic effect, slowing svRT.

Conclusions: Our findings highlight the positive impact whereby properly treating disease states, such as diabetes, can have on FH-associated risk, opening the door to the development of more targeted risk reduction approaches to combat AD.

**EFFECTS OF NPTX2 KNOCKOUT ON BEHAVIOR, BRAIN VOLUME BY MRI AND CA1 HIPPOCAMPAL SINGLE UNIT PROPERTIES.** Terrazas A, Pyon W, Zempare M, K. F. Young KF, Dalmendray A, Do L, David B, Bohne KM, Carey NJ, Chawla MK, Trouard TP, Zhou J, Worley PF, Barnes CA. University of Arizona; Johns Hopkins School of Medicine; Arizona Alzheimer's Consortium.

**Background:** Long-term memory is dependent on rapid, de novo protein synthesis, and we have studied immediate early genes (IEG) that act at excitatory synapses to understand the molecular basis of memory. IEGs Arc, Homer1a, and NPTX2 each play a unique and essential role in homeostatic restoration of activity levels of pyramidal neuron-inhibitory interneuron circuits in culture (1) (2) (3). Here, we examine NPTX2, which is induced by patterned activity that induces synaptic plasticity (4), is trafficked along axons, secreted from presynaptic elements, and selectively accumulates at excitatory synapses on PV-interneurons (3). This synaptic NPTX2 binds postsynaptic AMPAR (GluA1 and GluA4) and strengthens excitatory drive of PV-interneurons.

**Methods:** Sprague Dawley CRISPR-Cas9 NPTX2 knockout (KO) rats were compared to their wildtype (WT) littermates on 1) a battery of behavioral tasks (spatial and working memory on the watermaze, spontaneous object recognition memory and a test of anxiety), 2) MRI brain regional volumetric analysis, and 3) multiple single unit electrophysiological recordings in area CA1 of the hippocampus while rats traversed a 65 cm track for reward.

**Results:** The NPTX2 KO (n=7) rats exhibited similar performance to WT rats (n=5) on these tasks. Furthermore, regional brain volumes were not significantly different between KO and WT rats. Place-specific firing of CA1 single units, however, was strikingly different between groups. Compared to WT rats (n=2), KO rats (n=3) (8 to 15 mo) exhibited significantly lower spatial information content per spike and information per second (for KO and WT respectively, number of single units =142 and 63, mean information per spike +/- std = 0.66 +/- 0.52 and 1.10 +/- 0.72,  $t=-4.30$ ,  $p<0.0005$ ; mean information per second = 0.59 +/- .79 and 1.06 +/- 0.93,  $t=-3.48$ ,  $p<0.005$ .) Firing rates of pyramidal cells were assessed while rats were on the maze and during pre- and post-maze sleep periods. There was a trend for the firing rates of hippocampal place cells to be higher in KO rats for all behavioral conditions, and this reached statistical significance in post-behavior sleep sessions (mean rate in Hz 0.84 +/- 1.14 compared to 0.54 +/- 0.60 for WT,  $t=2.40$ ,  $p<0.05$ ). Surprisingly, the reduction of information content and spatial tuning of place cells is not manifest as deficits in the standard behavioral tasks used.

**Conclusions:** NPTX2 is required for developmental plasticity in the visual cortex to refine tuning properties of excitatory neurons (5), and a similar role in the hippocampus might create circuits capable of optimal information processing. In fact, it is possible that, during development, NPTX2 sets the spatial tuning parameter space for hippocampal networks.

**MODULATION OF ENDOTHELIAL CELL HYPOXIC INJURY BY AMYLOIDOGENIC MEDIN: IMPLICATIONS FOR AGING-RELATED CEREBROVASCULAR DISEASE.** Truran S, Karamanova N, Davies HA, Hansen M, Weissig V, Madine J, Migrino RQ. Phoenix Veterans Affairs Medical Center; Midwestern University; University of Liverpool; University of Arizona School of Medicine-Phoenix; Arizona Alzheimer's Consortium.

**Background:** Medin is a 50-amino acid amyloidogenic peptide that accumulates in the vasculature with aging. We recently showed that medin is present in cerebral arteries of elderly brain donors with greater amount in vascular dementia versus cognitively normal subjects. Medin was also shown to cause endothelial dysfunction and inflammatory activation, the former related to induction of oxidative stress. These findings suggest that medin may be an important mediator in cerebrovascular disease and vascular dementia. The effect of medin on endothelial cell (EC) function in setting of hypoxic injury is not known. Nanoliposomes are <100 nM phospholipid-containing particles. We recently showed that monosialoganglioside-containing nanoliposomes (GM1L) prevent medin-induced EC death likely through Nrf2-dependent upregulation of antioxidant stress response. We are testing the hypotheses that 1) medin aggravates EC hypoxic injury, 2) GM1L would prevent medin and/or hypoxic injury to ECs.

**Methods:** Recombinant medin was expressed in Lemo 21 (DE3) cells using pOPINS-medin and confirmed to have >95% purity by SDS-PAGE. GM1L was prepared from phosphatidylcholine, cholesterol and monosialoganglioside (molar ratios 70:25:5) using lipid hydration method. Primary human brain microvascular ECs (Lonza, passages 4-8) were exposed for 20 hours to 4 different treatments (vehicle control, medin 5  $\mu$ M, medin 5  $\mu$ M + GM1L 300  $\mu$ g/ml or GM1L 300  $\mu$ g/ml) under two aerobic conditions: physoxia (5% oxygen) or hypoxia (1% oxygen). Cell viability was assessed using calcein-AM fluorescence (based on principle of intact esterases in viable cells that releases fluorescent calcein) using flow cytometry (measurements expressed relative to control ECs exposed to room air). Gene expression of antioxidant enzymes heme-oxygenase 1 (HO1), NADPH quinone dehydrogenase (NQO1) and superoxide dismutase 1 (SOD1) were measured separately by qPCR on ECs.

**Results:** Compared to physoxia condition (97.7 $\pm$ 6.6% viability), hypoxia reduced EC viability by 18.8% (79.3 $\pm$ 2.5%,  $p$ <0.05), medin exposure reduced it by 32% (66.5 $\pm$ 2.3%,  $p$ =0.003) and combined hypoxia and medin by 50% (48.9 $\pm$ 1.9%,  $p$ <0.001). NLGM1 co-treatment restored EC viability in setting of hypoxia, medin or hypoxia+medin exposure. NLGM1 upregulated EC gene expressions antioxidant proteins HO-1, NQO1 and SOD1.

**Conclusions:** Medin exposure showed significant additive adverse effect on viability of human brain microvascular endothelial cells exposed to hypoxic insult compared to medin alone. Because medin is associated with aging vasculature, and aging is associated with ischemic injury from atherosclerotic cerebrovascular disease, medin may be an important modulator of injury in advanced cerebrovascular disease and a potential novel treatment target. GM1-containing nanoliposomes prevented endothelial cell hypoxic injury as well as medin-mediated cell injury, likely through effects on upregulation of antioxidant defense mechanisms. Further tests need to be performed to evaluate its potential as novel treatment approach in ischemic cerebrovascular disease.

**INTERACTIONS BETWEEN WEIGHT, ACTIVITY, AND APOE MUTATIONS IN MICE.**  
Velázquez A, Egbo D, Wiegand JP, Brinton RD. University of Arizona; Arizona Alzheimer's Consortium.

Background: Alzheimer's Disease (AD) is a progressive neurodegenerative disease characterized by cognitive impairment and abnormal protein aggregation. An individual's risk for developing Alzheimer's disease is a sum of genetic, environmental, and lifestyle risk factors. While the APOE $\epsilon$ 4 allele is the most prevalent genetic AD risk factor, it confers low risk of developing AD and may instead compound environmental and lifestyle risk factors.

Methods: Recent studies have identified an inexplicable weight loss prior to cognitive deficits and have implicated changes in hyperactivity (characterized by restless fidgeting) and impaired lipid metabolism. Characterizing and understanding the relationship between known genetic risk factors and the trajectory of weight loss could provide an accurate biomarker for disease onset. Accordingly, the goal of the current study was to investigate weight and hyperactivity in APOE $\epsilon$ 3/3, APOE $\epsilon$ 3/4, or APOE $\epsilon$ 4/4 mice.

Results: As predicted, both male and female APOE $\epsilon$ 4/4 mice weigh less than the control APOE $\epsilon$ 3/3 mice at 10+ months of age. Behavioral data from gait analyses and open field tasks suggest that APOE $\epsilon$ 4/4 have a higher average run speed and cadence (steps/second) than APOE $\epsilon$ 3/3 mice at 14 months of age.

Conclusions: These findings may suggest that weight loss may be a reliable biomarker and help identify environmental and lifestyle risk factors. Future experiments will focus on a subset of under-weight animals undergoing nutritional interventions, which studies suggest are the most at-risk individuals. Using weight as a biomarker for AD would significantly expand the therapeutic treatment window before significant neuronal damage has occurred.

**THE ALZHEIMER'S PREVENTION INITIATIVE (API) GENERATION PROGRAM: A GLOBAL PERSPECTIVE ON BEST PRACTICES FOR SITE SUPPORT AND ENGAGEMENT TO ACCELERATE PARTICIPANT RECRUITMENT.** Walsh T, Riviere ME, Tariot PN, Aldred M, Doak K, Liu F, Ricart J, Scholten I, Arbuckle J, Quinn M, Duff L, Smith M, Graf A, Langbaum JB. Banner Alzheimer's Institute; Novartis Pharmaceuticals; Novartis Pharma; 4Novartis Pharma; Black Swan Pharmaceuticals; Arizona Alzheimer's Consortium.

Background: The API Generation Program consists of two trials: Generation Studies 1 and 2. Generation Study 1 enrolled cognitively unimpaired APOE4 homozygotes (HMs) ages 60-75; Generation Study 2 enrolled cognitively unimpaired APOE4 carriers (HMs and heterozygotes (HTs), HTs had to have elevated brain amyloid). To address the unique challenges with recruitment of this population, we developed a multifaceted approach to maintaining engagement with sites and sharing best practices for recruitment among study site teams.

Methods: We designed a flexible menu of novel recruitment tactics and implemented strategies, some of which were country-specific, and collaborated with site teams on a regular basis. We conducted individualized "recruitment planning and implementation" calls with sites soon after they were activated, traveled to sites for in-person meetings, held regional and country-wide workshops and roundtable meetings, bi-monthly study coordinator calls, and monthly media training calls. In addition, we developed an online recruitment and retention toolkit to provide a one-stop shop for sites to review available, IRB approved materials that could be ordered and customized to meet their site-specific needs. In the US, we consulted with UsA2 for cultural competency to develop a tip sheet for minority community engagement, postcards, ads, and brochures specific to the Latino and African American communities near study sites.

Results: Recruitment was terminated in July 2019 after an early signal of mild worsening in some measures of cognitive function with umibecestat. At the time the program closed, 35,333 participants had come to a study site for genetic testing, 9,623 screened and 1,626 were randomized. 50% of activated sites had enrolled at least five participants between both studies. Despite the unprecedented challenges for recruitment of cognitively normal APOE4 HMs and HTs, we were on target to complete enrollment by end of 2019. Results from specific engagement and recruitment strategies, along with characteristics of high enrolling sites, will be presented.

Conclusions: Collaboration between study sponsor and site teams and the development of flexible recruitment plans and materials that can be adapted to meet sites' needs played a crucial role in the successful recruitment, enrollment, and retention of participants for the API Generation Program.

**LONGITUDINAL DATA IN PERIPHERAL BLOOD CONFIRMS PM20D1 IS A QUANTITATIVE TRAIT LOCUS (QTL) FOR ALZHEIMER'S DISEASE AND IMPLICATES ITS DYNAMIC ROLE IN DISEASE PROGRESSION.** Wang Q, Chen Y, Readhead B, Chen K, Su Y, Reiman EM, Dudley J. Arizona State University; Banner Alzheimer's Institute; University of Arizona; Translational Genomics Research Institute; Icahn School of Medicine at Mount Sinai; Arizona Alzheimer's Consortium.

Background: Genome wide genetic/epigenetic studies are revealing more risk loci associated with the Alzheimer's disease (AD). One recent analysis of human epigenome-wide association studies (EWAS) reports an association between variations in the gene PM20D1 and AD, showing hypermethylation in the brain tissues in patients with advanced-stage AD.

Methods: We used whole genome DNA methylation data collected from peripheral blood mononuclear cells (PBMCs) in a cohort (n=649) from the Alzheimer's Disease Neuroimaging Initiative (ADNI) and compared the DNA methylation level at baseline among different diagnosis groups. We also leveraged the longitudinal data up to 4 years, sampled at approximately 1 year intervals to model the alterations in the methylation levels detected at the differentially methylated regions (DMRs) to delineate the methylation change course over aging and disease progression. The dynamic pattern of DNA methylation level with disease progression at the PM20D1 locus in brain tissues were also modeled from the data in The Religious Orders Study and Memory and Aging Project (ROSMAP) Study cohort (n=740).

Results: When compared with controls, patients with mild cognitive impairment (MCI) consistently displayed promoter hypomethylation at reported methylation QTL gene locus PM20D1. This promoter hypomethylation was even more prominent in patients with mild to moderate AD. This is in stark contrast with previously reported hypermethylation in brain tissues in patients with advanced-stage AD. By leveraging longitudinal DNA methylation data, linear mixed-effects (LME) modeling for the unchanged diagnosis groups and U-shape testing for those with changed diagnosis (converters) indicate that, we observe initial promoter hypomethylation of PM20D1 during MCI and early stage AD, which reverses to eventual promoter hypermethylation in the transition to late stage AD. We also confirm this observation in ROSMAP cohort as neuropathology deteriorates. Correlation of DNA methylation between blood and brain tissues at PM20D1 promoter region also demonstrates the pattern is consistent across tissues.

Conclusions: Our results confirm that PM20D1 is an mQTL for AD and demonstrate that it plays a dynamic role at different stages of the disease. Further in-depth study is thus warranted to fully decipher its role in the evolution of AD, and potentially explore its utility as a blood-based biomarker for AD.

**ALLOPREGNANOLONE POTENTIATES BIOENERGETIC CAPACITY AND RE-STRUCTURES MITOCHONDRIAL RETICULUM IN NEURONS AND ASTROCYTES.** Wang T, Chen S, Mao Z, Brinton RD. University of Arizona; Arizona Alzheimer's Consortium.

Background: We previously demonstrated that the neurosteroid allopregnanolone (Allo) promotes neural stem cell regeneration, reverses neurogenic, metabolic and cognitive deficits and reduces Alzheimer's disease (AD) pathology in a mouse model of AD. To determine the cell-type specific mechanisms of Allo in regulating brain energy metabolism, we assessed the effect of Allo on mitochondrial bioenergetic profile and their morphological changes in rat E18 hippocampal neurons and astrocytes.

Methods: E18 rat hippocampal neurons were cultured for 10 days in neurobasal medium (NBM) with 2% B27 and then starved in 0.2% B27 / NBM for 4 hours before treatment with either 100nM Allo or 0.001% Vehicle overnight. E18 rat hippocampal astrocyte were cultured for 10 days in DMEM:F12(1:1) with 10% FBS and then starved in 10% Charcoal stripped-FBS / DMEM:F12 for 24 hours before treatment with 100nM Allo or 0.001% Vehicle overnight. Upon completion of treatment, both cell types were subject to morphological, biochemical and metabolic characterization of their mitochondrial phenotype.

Results: In primary hippocampal neurons, Allo treatment significantly reversed supplement depletion-induced deficits in mitochondrial respiratory capacity and dendritic morphology via Ca<sup>2+</sup> dependent signaling. In parallel, in E18 hippocampal astrocytes, Allo rescued serum depletion-induced decline in mitochondrial spare respiratory capacity through increasing mitochondrial biogenesis. Allo treatment reduced the population of less efficient swollen globule mitochondria in both neurons and astrocytes. Specifically, in astrocytes, Allo decreased the number of hyperfused tubule mitochondria and increased small globule mitochondria. The effect of Allo on re-structuring supplement depletion-induced mitochondrial reticulum in astrocytes was further supported by the restoration of the balance between Drp1 and Opa1 expression, which are key regulators for mitochondrial fission and fusion, respectively.

Conclusions: Outcomes of our findings further support the promising therapeutic effects of Allo against bioenergetic deficits and mitochondrial inefficiency that emerge in early phases of AD, with mitochondrial dynamics being a potential key targeting mechanism.

**DYNAMIC METABOLIC AGING OF THE FEMALE BRAIN DURING ENDOCRINOLOGICAL AND CHRONOLOGICAL AGING.** Wang Y, Shuang Y, Mishra A, Bacon E, Yin F, Brinton RD. University of Arizona; University of Southern California; Arizona Alzheimer's Consortium.

Background: Natural aging and the perimenopausal transition are associated with brain glucose hypometabolism and mitochondrial dysfunction in females. The bioenergetic crisis is also a hallmark of late-onset Alzheimer's disease (LOAD). Comprehensive understanding of the dynamic metabolic aging process in the female brain can shed light on potential prevention and interventions windows of opportunities to promote healthy aging.

Methods: To capture the natural aging process in the aging female brain, we used a rat model recapitulating fundamental characteristics of human menopausal transition. Hippocampal metabolomic and lipidomic analysis were conducted to detect metabolic changes across different aging stages. Hippocampal transcriptomic profiling was also done to identify underlying mechanistic pathways.

Results: We observed systematic bioenergetic dysregulation in the aging female brain, as well as alternations in key metabolic regulators. Using an unbiased, discovery-based metabolomic and lipidomic approach, we characterized the dynamic adaptation of aging female brain from glucose centric to utilization alternative fuel sources including amino acids, fatty acids, lipids, and ketone bodies, and finally to anaerobic glycolysis, during endocrinological and chronological aging. Transcriptomic profiling of bioenergetic gene networks were consistent with the metabolomic profiles.

Conclusions: Collectively, these data provide a detailed profile across transcriptomic and metabolomic systems underlying bioenergetic functions in brain and its relationship to peripheral metabolic responses. Translationally, these findings provide insights into the complex dynamics of chronological and endocrinological bioenergetic aging in female brain, and potential therapeutic windows of opportunity.

## **RELATIONSHIPS BETWEEN EPISODIC AUTOBIOGRAPHICAL MEMORY CONSTRUCTION AND ELABORATION AMONG YOUNG AND COGNITIVELY NORMAL OLDER ADULTS.**

Wank AA, Jarrett KL, Andrews-Hanna J, Grilli MD. University of Arizona; Arizona Alzheimer's Consortium.

Background: The retrieval of episodic autobiographical memories (EAMs) consists of an initial construction stage where autobiographical knowledge is explored, and a memory is selected based on goals of retrieval. Once identified, episodic details come together during elaboration to elicit a sense of mental time travel. Normal cognitive aging has been associated with a semantic shift in construction and elaboration, which puts forth the possibility that a set of cognitive processes guide the entire retrieval course. To address this idea, we investigated the connection between the mental process of navigating EAM construction and detailed elaboration in young and cognitively normal older adults.

Methods: Young and older participants were asked to retrieve EAMs via a novel application of the "think-aloud" paradigm. Each item was designated as either a directly retrieved memory (i.e., one that came to mind instantaneously in response to the cue word) or a generatively retrieved memory (i.e., different degrees of abstract autobiographical memories guided the construction process) by using an adapted scoring procedure to categorize the think aloud process. Participants chose a set of six constructed memories to elaborate in detail, which were scored using the Autobiographical Interview scoring procedure.

Results: In the entire sample, the correlation between the average number of internal details produced in elaborations was positively related to successful generative (i.e., retrieved an EAM), but not direct, retrieval rates. When we examined young and older adults separately, we found a similar pattern of results. Lastly, we replicated a robust finding in the literature where older adults produced fewer internal details than young adults during EAM elaboration.

Conclusions: These results suggest similar cognitive processes underlie both successful generative retrieval and episodic elaboration of EAMs, whereas direct retrieval may depend on different cognitive processes that are largely unrelated to those of elaboration. They also provide preliminary evidence that this connection exists in both young and older adult populations.

**THE LINK BETWEEN EMOTIONAL CONTROL BELIEFS AND HEALTH AMONG FAMILY CAREGIVERS.** Yoo J, O'Connors MF. University of Arizona; Arizona Alzheimer's Consortium.

**Background:** The challenges of caregiving for family members with Alzheimer's disease or related dementia (ADRD) can elicit a great amount of negative emotions on a daily basis. Cumulative negative emotional responses have been linked to poor health, putting caregivers at a greater risk of health problems. However, individual variance exists among ADRD family caregivers in their emotional responses to caregiving stressors. People differ in the extent to which they regulate negative emotions effectively, which mitigate the effects of external stressors on biological and behavioral pathways to health. Importantly, evidence suggests that beliefs that one can control his or her own emotions- termed as emotional control beliefs- contribute to actual success in emotion regulation. Given these links, it is important to examine whether emotional control beliefs can be a protective factor for stress-related health risks among caregivers.

**Methods:** Samples and procedure: We recruited 113 family caregivers of patients with ADRD residing in the U.S. Participants were recruited from various methods including social media, flyers, referral from neuro-clinics and community agencies. Eligibility criteria for participation were 1) identifying themselves as the primary person to take care of the ADRD patient, 2) having been in the caregiving role for at least 3 months, and 3) being able to read and answer an English survey. In addition, we recruited Anglo and Latino Americans for a different aim of the project that is not a scope of this abstract. Online survey was administered for the study.

**Measures:** Emotional control beliefs were measured for three negative emotions that are commonly experienced by caregivers: anger, anxiety, and sadness. Participants rated the extent to which they agreed with items like "I have little control over my anger [anxiety, sadness]." Because the factor analysis showed that the same items for different emotions were loaded on the same latent factor, the ratings across the three emotions were aggregated to create a composite score of emotional control beliefs. We also measured negative emotions in caregiving by assessing how frequently they experienced anger, anxiety and sadness due to caregiving during the past month. Three indicators of health were measured as outcome variables: self-reported health status (the RAND-12 inventory), depressive symptoms (C-ESD scale), and health behaviors (e.g., getting enough sleep) during the past month. For covariates, measures were obtained for demographic variables, general self-efficacy, caregiving characteristics (e.g., type of relationship with the care recipient, duration and hours spent in caregiving), and objective care-related stressors (e.g., the Revised Memory and Behavior Problems Checklist).

**Results:** First, we tested whether control beliefs predicted health controlling for covariates using multivariate linear regressions. Higher emotional control beliefs predicted fewer depressive symptoms,  $b = -2.9$ ,  $p = .001$ , better self-reported health status,  $b = 16.5$ ,  $p = .004$  and more health behaviors,  $b = .41$ ,  $p < .001$ . Next, we explored whether the associations between emotional control beliefs and health would be mediated by negative emotions in caregiving, using bootstrapping ( $n = 5000$ ). Negative emotions mediated the associations between emotional control beliefs and the two health outcomes: depressive symptoms and health status.

**Conclusions:** Emotional control beliefs predict lower depression and better self-reported health status and behavior among family ADRD caregivers. The associations hold after controlling for demographic variables, caregiving characteristics, and the level of objective stressors. The exploratory analysis showed that the associations between emotional control beliefs and depressive symptoms and health-status were mediated by negative emotions, suggesting that the beliefs may affect these health outcomes partly through decreasing the frequency of negative emotions elicited by caring a person with ADRD. The findings highlight the role of emotional control beliefs in health risks of caregiving. Future research should examine whether and how emotional control beliefs may buffer the negative impact of caregiving stressors on health.

**EFFECTS OF INDUCED HYPERTENSION IN MIDDLE AGED CYP1A1-REN2 TRANSGENIC RATS.** Zempare M, Carey NJ, Dalmendray A, Young K, Bohne KM, Do L, Trouard T, Mitchell KD, Chawla MK, Huentelman MJ, Barnes CA. University of Arizona; Tulane University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

Background: Hypertension is associated with an increased risk of cardiovascular disease (CVD) and cognitive decline in aging humans (Keenan et al., 2011) with the onset occurring around middle-age (Wilkie et al., 1971). While prior research has suggested an association between CVD and cognitive decline in the elderly (Haring et al., 2013), it is also critical to investigate how this dynamic may evolve from middle to older age. In this study, Cyp1a1-Ren2 xenobiotic-inducible transgenic rats were used to model the gradual rate and age-of-onset observed in humans. In these transgenic rats, Ren2 expression in the kidney is driven by the Cyp1a1 promoter, which is activated by ingestion of indole-3-carbinol (I3C), causing elevated kidney angiotensin levels, increased arterial pressure, and reduced renal hemodynamics (Mitchell et al., 2006).

Methods: Fifteen month old male rats were assigned to either control or treatment diet groups, and given a battery of behavior tests to establish baseline cognition measures. Following these tests, the treatment group received a diet with 0.015% I3C while control rats received a global 18% protein rodent diet. Post-treatment, the same behavioral battery was given to assess the effect of hypertension on cognition. Gradual onset of hypertension was confirmed through systolic and diastolic blood pressure changes. Postmortem heart and kidney analysis replicated and expanded on recent studies (Willeman et al., 2019). The behavioral battery includes spatial and cued versions of the Morris watermaze, spontaneous object recognition (SOR) and a delayed matching- to-place working memory task.

Results: Analysis of the hippocampal-dependent spatial watermaze, the perirhinal cortex-dependent SOR, and the prefrontal cortex-dependent working memory task, suggest that these hypertensive rats maintain high performance levels on each behavioral task. While the treated group in this study did show significant cardiac and renal end organ damage as in the Willeman et al. (2019) study, we did not replicate the impairment observed in spatial memory in this larger cohort.

Conclusions: Further studies, including those that assess vascular damage in relevant brain regions, are needed to determine if the molecular and cellular changes observed in these animals are similar to those seen in the peripheral vasculature. For example, the persistence of normal cognition after hypertension may be due to compensatory mechanisms such as local modulation of vascular tone and arteriole diameter by neurons and glial cells.

**NEURAL DEDIFFERENTIATION IN THE HUMAN HIPPOCAMPUS UNDERLIES SPATIAL MEMORY DEFICITS IN NORMAL AGING.** Zheng L, Ekstrom AD. University of Arizona; Arizona Alzheimer's Consortium.

Background: Both normal aging and Alzheimer's disease are frequently associated with a reduction in spatial memory. Profound spatial memory deficits may even serve as an early indicator of AD. The hippocampus, a structure that both shows age-related and pathological changes related to AD, also plays a critical role in spatial memory. One idea is that aging results in dedifferentiation, in other words, insufficiently differentiated neural (and cognitive) representations. This idea, however, has not been tested in the context of spatial navigation. Measuring the differentiation of neural patterns within the hippocampus could provide insight into spatial memory impairments noted with aging.

Methods: We built two virtual environments with each consisting of 6 stores. All participants (old adults and young adults) watched videos of two virtual environments on a computer to learn the locations of stores. Each city differed in the identity of the stores participants had to learn. Following learning, participants retrieved information about spatial distances while undergoing high-resolution functional magnetic resonance imaging (fMRI) targeting the hippocampus. To look at differentiation/dedifferentiation, we correlated voxel patterns within and between environments within an anatomically defined CA3/DG region of interest. Our protocol was similar to what we employed previously in Kyle et al. 2016 eLife with young adults except we included fewer spatial environments.

Results: Behavioral results showed worse spatial memory performance in older adults compared to younger adults. Using representational similarity analysis (RSA), in left CA3/DG, older and younger adults did not differ in the correlation of neural patterns within the same cities. In contrast, older adults showed higher across-city pattern similarity (PS) than younger adults. Further, we found that neural representations in older adults had greater variability, as measured by the standard deviation of MPS than in younger adults in CA3/DG.

Conclusions: These results provide novel evidence that the older adults show dedifferentiation in CA3/DG compared to younger adults during retrieval of different spatial environments. The unstable neural representations in CA3/DG, particularly between different environments, may be one of the reasons why the older adults show neural dedifferentiation compared to younger adults. Dedifferentiation in CA3/DG thus may provide a mechanistic account for spatial memory impairments in aging.

## **STUDENT ABSTRACTS**

## **EMOTION MATTERS: EPISODIC FUTURE THINKING IN YOUNG AND OLDER ADULTS.**

Acevedo-Molina MC, Novak A, Gregoire L, Mann L, Andrews-Hanna JR, Grilli MD. University of Arizona; Arizona Alzheimer's Consortium.

Background: The ability to construct hypothetical scenarios of events that pertain to our personal future is known as episodic future thinking (EFT) and is an essential part of our everyday lives. Prior research suggests that in young adults, positive and negative future events are constructed in richer episodic detail relative to neutral events. However, whether emotion also benefits the episodic specificity of future thinking among older adults is not known. The influence of emotion on the memorability of episodic future thoughts among older adults also has not been elucidated. Therefore, the aim of the present study was to investigate the effect of emotion on the specificity and memorability of EFT among young and older adults.

Methods: Forty cognitively and emotionally healthy adults (20 young and 20 older) selected and narrated 5 positive, 5 negative, and 5 neutral events that could happen to them 5 years in the future. They also rated the events for various phenomenological characteristics. After a 48-hour delay, participants were asked to remember and describe the future events they provided in the first session. The narratives were recorded, transcribed, and scored for episodic and non-episodic detail using the Autobiographical Interview protocol.

Results: For both young and older adults, positive future events were described with the most episodic detail, followed by negative future events, and then neutral future events. In both groups, more non-episodic details were also generated for emotional events (e.g. positive and negative) relative to neutral. In regard to the memorability of these events, both groups generated more episodic details for positive and negative events after a delay, relative to neutral events.

Conclusions: Future events that are imbued with emotion, positive or negative, are imagined with more episodic details by young and older adults. The emotion-related episodic detail advantage is heightened for positive future events. Additionally, more episodic details are generated for emotional events after a delay, suggesting that the effect of emotion carries over to one's memory for these future scenarios. Our findings suggest that despite there being an age-related reduction in episodic detail generation for simulating the future, older adults, similar to young adults, demonstrate an emotion-related boost in the episodic richness of EFT.

**MIND WINDOW: AN ACCESSIBLE MOBILE APP WITH PROMISE FOR REAL-WORLD MONITORING OF ALZHEIMER'S DISEASE RISK AND PROGRESSION.** Andrews EA, Mehl MR, Allen JJB, Griffiths C, Raffaelli Q, Grilli M, Andrews-Hanna JR. University of Arizona; Arizona Alzheimer's Consortium.

Background: Although conscious thought is an aspect of everyday life, relatively little is known about how thoughts manifest in the context that matters most: everyday life. Tracking "real-world" cognition could offer important insight into cognitive and affective mechanisms that shape healthy and pathological aging, including Alzheimer's disease. The ubiquity of smartphones provides a promising means for which to explore real-world cognition in large, demographically-heterogeneous samples over time.

Methods: To better understand the relationship between real-world thought and psychological health, we developed a freely-downloadable, cross-platform mobile application, called Mind Window, to solicit numerous characteristics of users' mood and thoughts over an indefinite period. Using a mobile application to sample introspective aspects of thought in the moment and within daily life allows for a highly intricate measure of subjective experience. In order to gather a broad description of thinking, questions sample: the manner of thought (intentionality, domain, and lability), content-related features, and associated affective qualities. In addition to longitudinal sampling, established measures will be used to assess trait characteristics.

Results: In this poster presentation, we will provide an overview of our app and report on preliminary findings from a pilot study in young and older adults. In this pilot study, data show that older adults experienced fewer real-world autobiographical thoughts, and rated their thoughts as more on-task, more positive, and more controllable. Additionally, factor analysis identified a latent variable that seemed to represent 'maladaptive repetitive thought' and which correlated with both increased depressive symptoms and poor psychological well-being.

Conclusions: If conscious, real-world thought can be characterized broadly, and specific patterns are shown to be related to psychological health, Mind Window may provide an accessible, non-invasive, and practical diagnostic tool to aid in identifying AD risk, onset, and disease progression. Such tools could ultimately provide a means for timely and targeted intervention – increasing psychological well-being and potentially reducing risk for age-related cognitive decline.

**TREATMENT OF PARKINSON'S DISEASE WITH ENHANCED DELIVERY OF ANTIBODY THERAPY SELECTIVELY TARGETING TOXIC PROTEIN VARIANTS.** Awasthi S, Murphy D, He P, Howison C, Erickson R, Matsunaga T, Trouard T, Sierks M. University of Arizona; Arizona State University; Arizona Alzheimer's Consortium.

**Background:** The onset and progression of Parkinson's disease (PD) has been linked to the formation and accumulation of toxic aggregates of alpha-synuclein (a-syn). Since the monomeric form of a-syn has beneficial functions for neurons, effective therapeutics for treating PD should selectively target only the toxic protein aggregates while leaving the beneficial protein forms untouched. We have developed antibody-based reagents that selectively target these toxic variants and have demonstrated that these antibody fragments have excellent therapeutic benefit when administered systemically in mouse models of PD.

**Methods:** Here we propose to further develop the most promising antibody reagents as a potential therapeutic for PD by administering the full-length antibody form of the selected reagent in a mouse model of PD. We will employ two different methods to increase delivery of the antibody therapeutic across the blood-brain barrier (BBB); 1) addition of a peptide tag to facilitate translocation of the antibody across the BBB; and 2) transient disruption of the BBB using a focused ultrasound (FUS) treatment. This proposal will enable us to demonstrate effectiveness of the therapeutic in a full length antibody format which will facilitate an investigational new drug application to the FDA. The antibody fragment has been expressed as a full length mouse IgG antibody for therapeutic trials in the mouse model. The antibody is administered by intraperitoneal injection. At the completion of the study, mice will be sacrificed and brain tissue analyzed for levels of oligomeric and fibrillar a-syn, brain inflammation, and neuronal integrity.

**Results:** In preliminary studies we have shown that the full length mouse IgG maintains binding specificity, and that it does cross the BBB with slightly better efficiency with FUS than without. We have also shown that the therapeutic IgG does successfully enter neurons in the mouse brain and successfully engages the toxic a-syn target. We are currently performing long term therapeutic studies to determine the therapeutic benefit of the IgG when administered systemically with and without FUS treatment.

**Conclusions:** We have developed an antibody based therapeutic that selectively targets toxic a-syn aggregates without blocking function of beneficial monomeric a-syn. We have previously shown that this antibody fragment provides excellent therapeutic benefit in a mouse model of PD. Here we show that the antibody fragment maintains its binding specificity when converted to a full length IgG format. We have shown that with a peptide tag to facilitate transport into and out of cells and FUS to increase transport across the BBB, that the IgG successfully engages the toxic a-syn targets in a mouse model of PD. We are currently performing longer term therapeutic studies in the PD mouse model.

**HEALTHY COGNITIVE AGING SHOWS DIFFERENTIAL IMPACTS ON DIRECTED AND RANDOM EXPLORATION.** Bauer K, Mizell JM, Wang S, Keung W, Alvarado L, Skupny AF, Frisvold A, Sundman MH, Alexander G, Wilson RC. University of Arizona; Arizona Alzheimer's Consortium.

Background: In making decisions, we all must weigh the advantages of exploring new but unknown options and exploiting familiar options. For instance, when purchasing running shoes, do you explore a new brand with few but promising reviews, or do you exploit the same shoes you have used in the past and know work for you? Older adults face important explore-exploit decisions in terms of retirement savings and health treatments. Previous research on younger adults has indicated that people use two strategies to solve these explore-exploit decisions: directed exploration and random exploration. Directed exploration is guided by information-seeking, or choosing an option that will reveal more information. Low willingness to seek information denotes higher ambiguity aversion. Random exploration is guided by random decision variability, or errors in the decision process.

Methods: This study uses an explore-exploit decision paradigm to measure these two types of exploration behavior in older adults. We constructed a task that manipulates information about each option available to participants and controls the number of choices in each trial, to measure directed and random exploration as well as ambiguity aversion and decision variability.

Results: Findings indicate that older adults show reduced random exploration and increased ambiguity aversion compared to young adults. We also find differences in these decisions between cognitively healthy older adults and those with cognitive impairments. Cognitively impaired older adults show no random exploration and higher baseline decision variability, plus a trend toward more ambiguity aversion.

Conclusions: Our results suggest that older adults could be missing out on favorable new options by avoiding seeking information, and cognitively impaired adults are random in their choices, so they may struggle to make educated decisions. Future research can examine how neuropsychological variables predict decision behavior, to eventually help adults maintain their ability to make informed decisions.

**THE IMPLICATION OF SRC FAMILY KINASES IN ALZHEIMER'S DISEASE.** Beirute-Herrera JL, Diouf D, Van den Hove DLA, Mastroeni D. Maastricht University; Arizona State University; Arizona Alzheimer's Consortium.

Background: Alzheimer's Disease (AD) remains as one of the major challenges for the neuroscientific community today. It affects approximately 50 million cases worldwide, with a total cost estimation of \$290 billion in 2019 (Alzheimer's Association, 2019). The pathophysiological hallmarks of AD are amyloid beta plaques (A $\beta$ ) and neurofibrillary tangles (NFTs). Nevertheless, the underlying mechanism have yet to be elucidated (Mehta et al., 2017). Recent efforts have demonstrated that non-receptor Src-family tyrosine kinases in AD are potential targets for therapeutic intervention and we show epigenetic mechanisms associated with the expression of non-receptor Src-family tyrosine kinase (e.g. methylation), as a mechanism for its dysfunction (Kaufman et al., 2015; Iatrou, Kenis, Rutten, Lunnon & van den Hove, 2017). The current project aims to investigate the proteomic, genomic and epigenomic correlation of c- and v-SRC to PYK2 and Fyn in neuropathologically confirmed AD and nondemented control cases.

Methods: Western blots, immunohistochemistry, qPCR and Methylation Specific PCR will be performed in the Medial Temporal Gyrus (MTG) of 10 AD cases in a range of disease states (Braak stage IV, V and VI) and in 10 healthy age matched controls (Braak stage 0-II). Laser Capture Microdissection will be used prior to qPCR to select neurons and microglia. For the statistical analysis, a 2-tail student t test and regression analysis of expression profiles will be conducted using IBM SPSS Statistics 25.0 with a significance level of >0.05 and covariates like Braak stage and gender.

Results: Preliminary results already showed a significant upregulation of v-Src and Fyn in CA1 laser capture neurons in AD cases compared to controls. While immunohistochemistry shows an upregulation of the Src family kinases in the CA1 of the AD cases. We hypothesize that there is an upregulation of c-Src, v-Src, Fyn and PYK2 mRNA/protein levels in the MTG of AD cases, which is correlated to hypomethylation in the promoter regions of the corresponding genes in AD cases. Furthermore, is expected that the same proteins interact in both microglial and neuronal cells following an equivalent pattern.

Conclusions: V-Src and Fyn are upregulated in Alzheimer's Disease cases and must be considered to further study the pathophysiology of the disease.

**SELECTIVE THALAMIC NUCLEI ATROPHY IN ALZHEIMER'S DISEASE: A NEUROIMAGING STUDY.** Bernstein AS, Rapcsak SZ, Saranathan M. University of Arizona; Arizona Alzheimer's Consortium.

**Background:** Until recently, the involvement of specific thalamic nuclei in Alzheimer's disease (AD) has remained largely uninvestigated, despite the well-known roles of the limbic thalamic nuclei, including the anterior ventral, anterior dorsal, lateral dorsal, and medial dorsal nuclei in episodic memory. In this work, a novel MRI-based imaging technique and thalamic segmentation method are used to investigate the involvement of thalamic nuclei in AD. Volumes of several thalamic nuclei are compared to obtain new insights into the pathogenesis of AD.

**Methods:** Thirteen healthy controls (HC), sixteen subjects with mild cognitive impairment (MCI), and 20 subjects with a clinical diagnosis of AD were included in the current study with IRB approval. All subjects were scanned using a white-matter-nulled (WMn) MPRAGE T1-weighted imaging sequence. Left and right thalami of each subject were automatically segmented using the THalamus Optimized Multi Atlas Segmentation (THOMAS) pipeline, to compute volumes of nuclei and the whole thalamus. Volumes were adjusted for total intracranial volume, determined using FREESURFER, and age for all subjects and nuclei. Thalamic volumes between HC and MCI as well as between HC and AD were compared using a student's t-test. Multiple comparisons were corrected using the false discovery rate (FDR) correction, and a significance level of  $p < 0.05$  was set for the corrected p-values.

**Results:** When comparing the HC and AD groups, there were significant differences in the total volume of the left thalamus ( $4797 \text{ mm}^3 \pm 525 \text{ mm}^3$  vs.  $4426 \text{ mm}^3 \pm 462 \text{ mm}^3$ ,  $p = 0.02$ ) and right thalamus ( $4861 \text{ mm}^3 \pm 387 \text{ mm}^3$  vs.  $4476 \text{ mm}^3 \pm 441 \text{ mm}^3$ ,  $p = 0.02$ ). There were also significant differences in the left and right ventral lateral anterior nuclei ( $76 \text{ mm}^3 \pm 15 \text{ mm}^3$  vs  $54 \text{ mm}^3 \pm 21 \text{ mm}^3$ ,  $p = 0.004$  on left,  $81 \text{ mm}^3 \pm 16 \text{ mm}^3$  vs  $58 \text{ mm}^3 \pm 23 \text{ mm}^3$ ,  $p = 0.01$  on right), the left and right pulvinar nuclei ( $1254 \text{ mm}^3 \pm 121 \text{ mm}^3$  vs  $1061 \text{ mm}^3 \pm 126 \text{ mm}^3$ ,  $p = 0.0004$  on left,  $1181 \text{ mm}^3 \pm 76 \text{ mm}^3$  vs  $1074 \text{ mm}^3 \pm 146 \text{ mm}^3$ ,  $p = 0.03$  on right), and the left and right medial geniculate nucleus ( $61 \text{ mm}^3 \pm 5 \text{ mm}^3$  vs  $50 \text{ mm}^3 \pm 7 \text{ mm}^3$ ,  $p = 0.0003$  on left,  $61 \text{ mm}^3 \pm 8 \text{ mm}^3$  vs  $54 \text{ mm}^3 \pm 6 \text{ mm}^3$ ,  $p = 0.02$  on right). When comparing the HC and MCI groups, there is a significant difference bilaterally in the ventral lateral anterior nuclei ( $76 \text{ mm}^3 \pm 15 \text{ mm}^3$  vs  $52 \text{ mm}^3 \pm 19 \text{ mm}^3$ ,  $p = 0.003$  on left,  $81 \text{ mm}^3 \pm 16 \text{ mm}^3$  vs  $51 \text{ mm}^3 \pm 19 \text{ mm}^3$ ,  $p = 0.0003$  on right).

**Conclusions:** This preliminary work demonstrates for the first time that several thalamic nuclei preferentially atrophy in AD, including the left and right ventral lateral anterior nuclei, the left and right pulvinar nuclei, and the left and right medial geniculate nucleus. In addition, the left and right ventral lateral anterior nuclei show a significant decrease in volume in subjects with MCI. While this work is preliminary and requires more data, it suggests that further investigation of the thalamus' role in AD may be warranted. Detectable changes in the ventral lateral anterior nucleus in subjects with MCI also suggests that this nucleus may be affected early in the progression of the disease and could serve as a target for early diagnosis.

**ALZHEIMER'S DISEASE FLUID BIOMARKERS RELATED GRAY MATTER COVARIANCE PATTERNS IN HEALTHY OLDER ADULTS.** Bharadwaj PK, Andrews-Hanna JR, Kuo P, Alexander GE. University of Arizona; Banner University Medical Center Tucson; Arizona Alzheimer's Consortium.

Background: Biomarkers of Alzheimer's disease (AD) pathology in cerebrospinal fluid (CSF), including A $\beta$ 42, p-Tau181, and the ratio of p-Tau181/A $\beta$ 42, can help identify those with increased risk for dementia, before the onset of clinical symptoms. How these CSF biomarkers relate to regional patterns of gray matter (GM) atrophy in cognitively unimpaired older adults has yet to be fully investigated.

Methods: Here, we applied a multivariate network analysis technique, the scaled subprofile model (SSM; Alexander & Moeller, 1994) to identify gray matter covariance patterns related to CSF measures of A $\beta$ 42, p-Tau181, and the ratio of p-Tau181/A $\beta$ 42, in a sample of healthy older adults drawn from the Alzheimer's Disease Neuroimaging Initiative (ADNI2; N=146; Age=73.5  $\pm$  6.4 years, range=56-89 years; sex (F/M)=76/70; Education= 16.5  $\pm$  2.5 years; MMSE=29.1  $\pm$  1.2; CDR = 0; APOE- $\epsilon$ 4 (N/Y)=108/38). GM maps were segmented from 3T T1-weighted volumetric magnetic resonance imaging (MRI) scans with SPM12, spatially normalized using diffeomorphic registration (DARTEL), and smoothed with a Gaussian kernel of 10mm. Regional SSM network analysis was performed on these GM maps using Akaike Information Criteria with 2000 Bootstrap iterations to identify linear combinations of GM patterns associated with each of the three fluid biomarker measures.

Results: The p-Tau181/A $\beta$ 42 - related GM SSM pattern ( $R^2 = 0.10$ ,  $p \leq 4.4E-05$ ) was characterized by bilateral reductions in the vicinity of the superior temporal gyrus (STG), and extensive bilateral GM reductions in cerebellar lobules. The p-Tau181 related GM SSM pattern ( $R^2 = 0.06$ ,  $p \leq 2.75E-03$ ) showed extensive bilateral reductions in the cerebellum extending into the anterior cerebellar lobule. In contrast, CSF A $\beta$ 42 did not exhibit an SSM pattern with robust regional GM contributions in this cohort. Additionally, the p-Tau181/A $\beta$ 42 ( $R^2$  change = 0.042,  $p$ -change  $\leq 0.007$ ) and p-Tau181 ( $R^2$  change = 0.042,  $p$ -change  $\leq 0.008$ ) related GM patterns each remained significantly associated with their respective CSF biomarker, after adjusting for age, sex, years of education, APOE genotype, hypertension status, and intracranial volume. Greater expression of the p-Tau181/A $\beta$ 42 - GM pattern was associated with lower scores on the Clock Drawing Test ( $R^2$  change = 0.047,  $p$ -change  $\leq 0.007$ ) with the same set of covariates.

Conclusions: These results demonstrate that AD-related CSF biomarkers including p-Tau181 and its ratio to A $\beta$ 42 may be associated with individual differences in regional topographies of GM volume, involving temporal and cerebellar brain regions, in healthy cognitively unimpaired older adults. Together, these findings provide further support for the use of CSF fluid biomarkers in evaluating the earliest preclinical effects of AD and their relation to the effects of brain aging.

**INFLUENCE OF FECAL MICROBIOTA TRANSPLANTATION ON NEUROINFLAMMATION OF TRIPLE TRANSGENIC AD MICE.** Borsom EM, Keefe CR, Hirsch A, Orsini GM, Conn K, Jaramillo SA, Kask O, Bolyen E, Dillon M, Lee K, Caporaso JG, Cope EK. Northern Arizona University; Arizona Alzheimer's Consortium.

**Background:** The human microbiota, the aggregate of all bacterial, viral, fungal, and archaeal cells that inhabit the human body, contribute to diverse aspects of human health, including immune function, protection against pathogen colonization, and metabolism. The gut microbiota bidirectionally communicates with the brain via cytokines, neurotransmitters, hormones, and secondary metabolites, known collectively as the gut-brain axis. The gut-brain axis is suspected to be involved in the development of neurological diseases, including Alzheimer's disease (AD), Parkinson's disease, and Autism spectrum disorder. AD is characterized by plaque deposition, tau tangles, and neuroinflammation. Individuals differ widely in the composition of their gut microbiota. In preliminary results, we found microbial gut communities were compositionally distinct in 3xTg-AD mice. We hypothesize that certain compositions of the gut microbiota, via the gut-brain axis, contribute to the development of AD pathologies and neuroinflammation. To further elucidate the role of the gut-brain axis in AD, we performed fecal microbiota transplants from aged, 3xTg-AD mice to pre-AD, 3xTg-AD mice to shift the gut microbiota towards an AD-like state, in attempt to increase the rate of pathogenesis. If successful, we will have identified a gut microbiota composition that contributes to AD development, and we will use that to understand what features of the gut microbiota may be involved in AD development.

**Methods:** Fecal samples were collected from mice fortnightly and fecal microbiota transplants (FMT) were performed via oral gavage, transplanting a fecal slurry (half fresh, half frozen) from aged, 3xTg-AD mice directly into the gut of young, pre-AD 3xTg-AD or wildtype (WT) mice. At 8 weeks, FMTs were performed for 5 consecutive days, followed by fortnightly maintenance transplants. DNA was extracted from fecal samples, the V4 region of the 16S rRNA gene was amplified, and amplicons were sequenced on the Illumina MiSeq. Data were analyzed using QIIME2. Additionally, a custom qPCR assay was used to assess inflammation in the hippocampus of the FMT cohort at 24 weeks of age. The assay was comprised of various biomarkers for microgliosis/M1/Th1 (iNOS, IFN-gamma, IL-1B, IL-12B, IL-2, CCL2) and M2/Th2 (arg-1, IL-4, IL-5, IL-6, IL-10, TNF-alpha) inflammation.

**Results:** Our results show a shift in microbiome composition in FMT-positive mice when compared to FMT-negative mice. The key genera involved in this shift are Bacteroides and Prevotella. In 3xTg-AD and WT mice, there was no difference in neuroinflammation between FMT and PBS mice.

**Conclusions:** The gut microbiota composition in FMT-treated mice shifted with engraftment of aged 3xTg-AD gut microbial communities. 3xTg-AD and WT mice who received FMTs from aged 3xTg-AD had no significant change in neuroinflammation.

**ESTROGEN MODULATING BREAST CANCER THERAPIES AND THE INCIDENCE OF ALZHEIMER'S DISEASE.** Branigan GL, Soto M, Neumayer L, Rodgers K, Brinton RD. University of Arizona; Arizona Alzheimer's Consortium.

Background: Globally, neurological disorders rank as the leading cause of disability-adjusted life-years, and the second-leading cause of deaths. The high global prevalence and economic impact of Alzheimer's disease presents a significant public health challenge while the identification of therapies to prevent Alzheimer's disease (AD) remains a challenge. Worldwide, breast cancer is the most common non-skin cancer in women. Approximately 1 in 8 women will be diagnosed with breast cancer during their lifetime. Currently, Alzheimer's disease affects 1 in 9 persons in the US over the age of 65, two thirds of whom are women.

Methods: The objective of this study was to determine whether exposure to hormone modulating therapies targeting estrogen for the treatment of breast cancer impact the risk of AD using medical informatics and the Symphony claims dataset. The Symphony dataset contains claims from private-payer and Medicare insurance datasets from across the United States. Survival analysis was used to determine the association between estrogen modulating therapy exposure and diagnosis of AD in the post index date follow-up period. A propensity score approach was used to minimize measured and unmeasured selection bias.

Results: In this cohort study of propensity score matched perimenopausal to postmenopausal aged women with breast cancer, estrogen modulating therapy exposure was associated with decrease in diagnosis of neurodegenerative disease, most specifically Alzheimer's disease. Kaplan-Meier curves for AD-free survival were analyzed to measure the overall rate and proportion of patients in each treatment group who went on to develop neurodegenerative disease.

Conclusions: Among female patients with breast cancer, exposure to estrogen modulating therapies, specifically tamoxifen and steroidal aromatase inhibitors, was associated with a decrease in diagnosis of AD and Dementia. As a result, translational studies investigating the mechanisms by which these therapeutics impact AD risk are currently underway.

**DEFICITS IN SPATIAL WORKING MEMORY DEPEND ON AGE IN A NOVEL RAT MODEL OF ALZHEIMER'S DISEASE.** Bulen HL, Woner VE, Northup-Smith SN, Peña VL, Manzo A, Valenzuela Sanchez M, Koebele SV, Bimonte-Nelson HA. Arizona State University; Arizona Alzheimer's Consortium.

**Background:** There are currently no disease-modifying treatments to halt or attenuate the progression of Alzheimer's disease (AD). Transgenic rodent models have provided researchers the ability to recapitulate particular pathological and symptomological events in disease progression. However, complete reproduction of all features of AD in an animal model has not yet been achieved, potentially leading to the inconclusive treatment results that typically occur at the clinical level. Recently, the TgF344-AD transgenic rat model, expressing the mutant human amyloid precursor protein (APPSW) and presenilin 1 (PS1 $\square$ E9) genes, has been developed, and has been shown to exhibit beta-amyloid-like pathology, tau hyperphosphorylation, and neuronal loss. However, this model has not yet been well-characterized in terms of its cognition across the lifespan, which is fundamental to understanding the trajectory of aging relative to pathology and learning and memory changes. We were particularly interested in evaluating female rats, as women constitute approximately 2/3 of clinical AD cases. Further, we aimed to characterize learning and memory in adulthood, during a potential "prodromal" phase of disease development, to assess whether cognitive impairments are evident prior to extensive neuropathology development.

**Methods:** The current project utilized the TgF344-AD rat model to systematically characterize spatial learning and memory performance in 60 female transgenic (Tg) and wildtype (WT) rats at 6, 9, and 12 months of age. Six groups were included in this study: 6 mo-Tg (n=10), 9 mo-Tg (n=11), 12 mo-Tg (n=9), 6 mo-WT (n=10), 9 mo-WT (n=10), and 12 mo-WT (n=10). All animals were bred, aged, and then tested during the same behavioral assessment timeframe. The behavioral battery consisted of the water radial-arm maze (WRAM) to assess spatial working and reference memory, the Morris water maze (MWM) to assess spatial reference memory, and the visible platform (VP) task to assess motor and visual acuity for performing a water-escape task. WRAM will be presented in this poster; other behavioral data and neuropathology are currently being processed and quantified.

**Results:** Preliminary results indicate that during the Learning Phase of WRAM, 6 mo-Tg rats tended to have impaired working memory compared to 6 mo-WT controls. This impairment was not evident during the Learning Phase for the 9 mo or 12 mo group comparisons. In the Asymptotic Phase of the WRAM, 6 mo-Tg rats tended to exhibit impaired working memory compared to 6 mo-WT rats, and 12 mo-Tg rats had significantly impaired working memory compared to 12 mo-WT rats. Interestingly, the 9 mo-Tg rats did not demonstrate a significant difference in working memory errors compared to the 9 mo-WT rats during the Asymptotic Phase of WRAM.

**Conclusions:** These results provide evidence for changes in spatial working memory performance as early as young adulthood, as well as yield insight into how these cognitive symptoms progress with aging in the TgF344-AD model. Further, this pattern of impairment, wherein Tg animals made more working memory errors compared to WT animals at the 6 and 12 month time points, but not at the 9 month time point, may be indicative of a transient compensatory behavioral response in adulthood that proves helpful at incipient stages of disease progression but eventually leads to exacerbated cognitive impairment by middle-age. Moreover, the preliminary findings from this study corroborate emerging literature suggesting that spatial working memory impairment may be an early behavioral marker of AD and should be systematically assessed at the clinical level. Future directions include analyses of the MWM to understand if the same pattern of impairment extends to pure spatial reference memory, as well as analyses of AD-like brain pathology, which will be used to glean insight into the temporal progression of pathological and cognitive outcomes

**MECHANISMS OF NEUROPROTECTION IN MAS AGONISTS USING A NOVEL VASCULAR DEMENTIA MODEL.** Butler R, Soto M, Rodgers KE. University of Arizona; Arizona Alzheimer's Consortium.

Background: Alzheimer's (AD) is a debilitating disease characterized by a loss of memory and executive function due to an increase in cytotoxic A $\beta$ . A growing number of studies have shown a direct correlation between hypertension and an increased risk of AD like symptoms, including cognitive deficits and increased A $\beta$  production. Furthermore, Renin-Angiotensin System (RAS) targeted anti-hypertensives have been shown to reduce the risk of AD development. This prompted studies that have demonstrated that the protective arm of RAS, angiotensin (1-7)[A(1-7)]/MAS receptor/angiotensin converting enzyme 2, is reduced in AD patients and that Mas activation increases cognition and is cardioprotective. Thus, we hypothesized that MAS agonists could help ameliorate AD-like pathology in mice using a mouse model of heart failure – transverse aortic constriction (TAC).

Methods: One week after induction of TAC in 8-10wk male C57Bl/6J mice, animals started treatment with saline or one of four Mas agonists. Four weeks after surgery, the cardiac function was assessed by echocardiography followed by assessment of their cognitive abilities (10 weeks) using novel object recognition. Upon necropsy at 90days, brain, heart, bone marrow and blood were collected and analyzed for phenotypic and functional changes.

Results: TAC surgery induced marked dysfunction in both standard cardiac measures of left ventricular function and cognition, which were rescued by MAS agonists treatment. MAS agonist also prevented an increase in antigen presenting microglia, thought to be the primary driver of AD pathology. This treatment effect was determined to be due to a protection of the mitochondrial oxidative phosphorylation pathway rather than biogenesis, and thus a reduction in mitochondrial dysfunction; in TAC mice, occurring more significantly in microglia and neurons. This trend was extended to the heart, improving outcomes for endothelial cells and cardiomyocytes. Lastly, TAC surgery affected viability of astrocytes and endothelial cell types in the brain, showing significant reduction in the total percentage, that was rescued with treatment.

Conclusions: The Mas agonist RASRx1902 was able to significantly decrease several measures of AD-like pathology in the TAC model, indicating that it might be effective in decreasing disease state in those with hypertension as risk factor for AD development. Future work will be focused on further characterizing treatment effects of RASRx1902 in biochemical measures of mitochondrial function, cellular pathway changes and evaluating efficacy of a concurrent pilot study examining cerebral blood flow and blood brain barrier integrity in TAC.

**AN MRI MICROSCOPY TOOLKIT FOR BRAIN MICRO-STRUCTURAL CHANGES IN AGING.**  
Comrie CJ, Dieckhaus L, Barnes CA, Gray DT, Hutchinson EB. University of Arizona; Arizona Alzheimer's Consortium.

Background: Developing tools that are sensitive to pathologies in aging, and age-related diseases such as Alzheimer's disease (AD) is critical to understanding normal and pathologic brain aging. Magnetic Resonance Imaging (MRI) microscopy is the scanning of fixed tissue at high-resolution. Quantitative MRI techniques give insight into microstructure and macromolecular alterations present in tissue environment. We developed a quantitative MRI microscopy battery that utilizes diffusion MRI, relaxometry, and magnetic susceptibility techniques for quantifying brain microstructure.

Methods: We optimized and applied Diffusion Tensor Imaging (DTI), selective inversion recovery (SIR) for T1 and bound pool fraction (BPF) mapping and multi-spin echo (MSE) for T2 and myelin water fraction (MWF) mapping in a fixed Bonnet macaque brain to evaluate image quality and to compare metric maps across techniques. We acquired DTI using 3D Echo Planar Imaging (EPI) at 500 micron isotropic with low b-value 5 shell (100-1200) and high b-value 2 shell (1500-3000). SIR and MSE were acquired using a 3D RARE pulse sequence. Data was processed offline using the UA HPC clusters. Raw SIR and MSE images required no artifact correction, but DTI had expected geometric distortions. DTI images were processed with TORTOISE: importing raw data directories, correcting motion artifacts, and EPI distortion. Final results received from the TORTOISE pipeline were Fractional Anisotropy (FA) and Diffusion-Encoded Color (DEC) maps. Both SIR and MSE were processed with the REMMI toolkit in MATLAB. REMMI toolkit outputted T1 weighted images and bound pool fraction map (BPF) from SIR pulse sequence, and T2-weighted images with Myelin Water Fraction map (MWF) from MSE.

Results: We have acquired DTI and REMMI on the Bruker 7T through Translational Bioimaging Resource on a normal aged Bonnet macaque brain (23 years old) provided by the Barnes' lab. Image resolution for all maps was high (400-500  $\mu\text{m}^3$  voxel resolution). For DTI, even with the presence of ghosting, we were able to obtain high quality diffusion maps and DWIs sufficient for performing accurate tractography maps. Acquired raw images from REMMI had high SNR and both BPF and MWF quantitative maps provided reasonable values in expected anatomical locations (e.g. white matter with high BPF and MWF). However, the BPF maps were more robust than MWF suggesting that the MSE fitting was not optimal and that improvement of MWF maps by acquisition or model development is needed.

Conclusions: The MRI microscopy battery was optimized and collected data on the healthy-aged Bonnet macaque brain and showed promising capabilities for the detection of tissue microstructural features. In the future, the battery will be used on more Bonnet macaque brains, all of which have undergone behavioral and electrophysiological assessments of cognitive and sensory function. The goal of these analyses is to understand how MRI outcome measures from distinct sensory and higher-order associative brain regions map onto brain function during healthy aging. Additionally, the lab will apply the battery to post-mortem human brains with healthy and Alzheimer's diseased models to identify quantitative markers of pathology by these MRI microscopy techniques.

**SPATIAL EYE-BLINK LEARNING BUT NOT AGE PREDICTS THETA-GAMMA COUPLING IN THE CA1 REGION OF THE HIPPOCAMPUS.** Crown L, Gray DT, Schimanski LA, Barnes CA, Cowen SL. University of Arizona; Simon Fraser University; Arizona Alzheimer's Consortium.

Background: Cross-frequency coupling (CFC) between theta- and gamma-band activity in the hippocampus has been linked to memory encoding and retrieval. Recent data suggest that the frequency of the gamma-band component of theta-gamma CFC differs as the relative contribution of entorhinal or hippocampal CA3 drive to mnemonic circuits within CA1 changes. While there are differing interpretations with respect to the generation of low gamma (25 - 50 Hz), one hypothesis is that it reflects drive from CA3 to CA1 while high gamma (75 - 90 Hz) denotes drive from medial entorhinal cortex (MEC) to CA1 (Colgin et al., 2009). Little is known about how activity in these frequency bands and their coupling changes with age; however, there are fewer functional Schaffer collateral synapses onto CA1 pyramidal cells and CA3 pyramidal cells show increased excitability. Given such alterations in these network properties, we hypothesized that high-gamma CFC associated with entorhinal input would be greater in aged animals.

Methods: To examine this question, local field potential activity of 12 rats (n = 6 young, 9-12 mo, n = 6 old, 25-28 mo) was analyzed as they performed a spatial eye-blink conditioning task (Schimanski et al., 2013). We measured low-gamma power, high-gamma power, and theta-gamma phase-amplitude coupling (PAC) as animals approached and departed from the region of the maze associated with a brief eyelid stimulation.

Results: We observed no difference between young and old animals in 1) peak gamma frequency, 2) in CFC, or 3) the ratio of low- to high-gamma power. Interestingly, we observed that animals that did not develop reliable eye blink conditioning (n = 5), regardless of their age, showed greater low-gamma relative to high-gamma power than those (n = 7) that did consistently show conditioning (two-sample t-test, p = 0.01). This effect was apparent after 5 days of training, suggesting that eye blink training altered the relative contribution of entorhinal drive to CA1. In addition, low-gamma PAC, but not high-gamma PAC, recorded as animals approached the eye-shock zone, was positively correlated with eye-blink learning in those animals that learned the task (one-sample t-test, p < 0.01), but not in animals that did not display learning, regardless of age.

Conclusions: Taken together, these results suggest that age is a less important predictor of CA1 theta/gamma dynamics than is performance

**AMYLOID  $\beta$  ANALYSIS FROM MICRODISSECTED BRAIN CELLS USING MICROFLUIDICS AND MALDI MASS SPECTROMETRY.** Cruz Villarreal J, Egatz-Gomez A, Coleman PD, Ros A. Arizona State University; Arizona Alzheimer's Consortium.

Background: While amyloid  $\beta$  ( $A\beta$ ) peptide plays a major role in Alzheimer's disease (AD) as one of the main constituents of brain plaques, soluble monomers to dodecameric oligomer  $A\beta$  species are involved in the complex pathology of the disease. Although a wealth of data is available about  $A\beta$  from in vitro studies, animal models and human brain homogenates, little is known about intracellular  $A\beta$  species in specific cell subpopulations, mainly due to the lack of analytical methods to access the proteomic content from selected brain cells. Identification of AD-related  $A\beta$  species in brain cell subpopulations could provide valuable insights on understanding the causes of AD. Thus, we propose to investigate the differences in  $A\beta$  species from AD and non-AD human brains. To address this interrogation, we develop a microfluidic-based assay for in-situ mass spectrometry (MS) analysis of  $A\beta$  from microdissected human brain cells.

Methods: To collect brain cells directly into a device, we couple laser microdissection (LCM) with our previously integrated microfluidic platform with matrix-assisted laser desorption ionization MS (MIMAS) for in-situ proteomic studies. All collection, handling and preparation steps can be performed on-chip, including proteomic content extraction, immunocapture of the peptide and addition of matrix solution for co-crystallization with the MALDI matrix. To characterize model oligomeric  $A\beta$  species synthesized in vitro, we explored different analytical techniques such as gel electrophoresis, atomic force microscopy, and MALDI-MS. To optimize the sample and matrix conditions for optimum MALDI ionization efficiency and sensitivity, we have further developed a microfluidic gradient generator to screen multiple conditions simultaneously.

Results: The MIMAS platform has been coupled with the LCM instrument, allowing to capture cells directly into microfluidic wells for further proteomic extraction. The immobilization of  $A\beta$  antibodies and immunocapture of the peptide on-chip have been successfully demonstrated. The synthesis of oligomeric  $A\beta$  species in vitro has been confirmed using different analytic techniques. A microfluidic device to optimize the sample-matrix ratio for MALDI-MS has been developed and tested.

Conclusions: The successful integration of the LCM-MIMAS approach will overcome the current limitations of accessing intracellular protein content in specific subpopulations, allowing the study of  $A\beta$  in cells from brain tissue. The immunocapture of  $A\beta$  has been shown by MALDI-MS using the MIMAS device. The preliminary results of  $A\beta$  characterization provide information to optimize the sample preparation and analysis conditions prior to  $A\beta$  analysis from brain tissue. A microfluidic device to screen multiple sample-matrix conditions has been developed and will further be explored for optimized MIMAS sensitivity. The resulting findings can provide valuable insights into the role of  $A\beta$  in AD progression.

**QUANTITATIVE MRI ANALYSIS OF RODENT BRAINS AS A FUNCTION OF AGE AND COGNITION.** Do L, Bernstein AS, Bharadwaj P, Ugonna C, Zempare MA, Chen N, Alexander GE, Barnes CA, Trouard T. University of Arizona; Arizona Alzheimer's Consortium.

Background: MRI, being both non-invasive and inherently translational, can play an important role in comparing brain anatomy in animal models and humans, and atlas-based tools for neuroinformatics can be used to study age dependent changes in brain anatomy and function. The work herein presents initial results from a large cross-sectional study employing a rodent model of aging to investigate the neuroanatomical and epigenetic correlates of healthy cognitive aging. Initial analysis of MRI data has utilized a rat brain template and associated atlas for comparison of rodent brains at various ages

Methods: Male Fisher 344 rats (n=114) were acquired at young adult (6 months, n=48), middle aged (15 months, n= 38) and old adult (23 months, n= 28) ages. These animals underwent a battery of behavioral tasks resulting in each age group being sub-divided into 3 sub groups of high cognition, average cognition and low cognition. Body weights were measured and neurological MRI was carried out on a 7T Bruker Biospec (Bruker, Billerica, MA). T2-weighted data, among other MRI scans, were collected with 150 $\mu$ m isotropic resolution. Images underwent brain extraction using a semi-automated process as well as bias correction due to non-uniform surface coil sensitivity using the ANTs software. A Waxholm Space Sprague Dawley T2-weighted template image registered to each individual animal in the study using linear and non-linear registration. The deformation fields produced from the registration were then applied to labeled atlas (80 regions of the brain) (1) using an in house Matlab code to calculate the volume of individual regions of interest (ROIs) in the brain. To compare ROI volumes across age and cognitive score, the volume of each ROI was normalized to total intracranial volume (TIV).

Results: TIV calculated from semi-automatic brain extraction are plotted for each age group shows significant difference from young adult to middle age and young adult to old adult, however there is no significant difference between middle aged and old adult groups. Body shows a significant increase from young adult to middle aged as well as old adult. However, there is a significant decrease from middle aged to old adult. The decrease in body weight from middle aged to old, while significant, was not substantial enough to bring the body weights back to the level of young adults. Total ventricular system volume and hippocampal volumes showed no significance across age or cognition.

Conclusions: For group-wise analysis considering different ages, total brain volume can be a confounding variable that needs to be accounted for. The findings in this study indicate that rat brains are growing from young adult to middle aged and plateaus through old age. Additionally, there was significant body weight increases from young adult to middle aged groups. This trend, however, did not persist as the bodyweight decreased significantly from middle aged to old adult. The total ventricular system was a region of particular interest because in human studies it has been shown to be an early indicator of aging and pathology. No significant differences were observed in TVS volume throughout the age or cognition. Body weight measurements confirm the imaging findings to middle aged however there is a deviation at old age where TIV volume plateaus and body weight decreases significantly. These results will inform future analysis comparing regional brain volumes with age and cognition.

**ARE YOUNG/OLD AGE-GROUP DIFFERENCES IN DECISION-MAKING BEHAVIOR MEDIATED BY CRYSTALLIZED OR FLUID INTELLIGENCE SCORES?** Duby N, Mizell J-M, Wang S, Keung W, Alvarado L, Farrell Skupny A, Frisvold A, Sundman MH, Alexander GE, Wilson RC. University of Arizona; Arizona Alzheimer's Consortium.

Background: Throughout our lives, we often face decisions in which we must trade off the relative benefits of exploring options that are unknown and exploiting options we know well. For example, when dining at our favorite Singapore-style restaurant, should we explore the unknown Fish Head Curry, or exploit the Rice Noodles we know and love? As we age, such explore-exploit choices take on increasing importance as we decide how to invest savings in retirement (e.g. explore a new balance of stocks and bonds or exploit the balance we've used so far?), or how to treat a chronic disease (e.g. explore new treatments with unknown side effects, or exploit known treatments we've used for years?). Aging is also associated with a reduction in some cognitive abilities such as executive functioning, processing speed, and episodic memory, collectively referred to as "Fluid Intelligence". Aging is also associated with preservation, or increase, in other abilities related to having more experience in the world, such as vocabulary, which are collectively referred to as "Crystallized Intelligence". The goal of the current preliminary study was to determine if these age group differences in decision-making behavior are mediated by crystallized or fluid intelligence scores.

Methods: To assess the constructs of fluid and crystallized intelligence, participants in the young and old age group underwent a battery of neuropsychology testing including the WAIS-IV intelligence test, Mini-Mental State Exam, North American Reading test, and others. We then scored the neuropsychological battery responses of participants and assessed their scores on various measures in order to determine their fluid and crystallized intelligence. To assess an individual's decision-making skills, we used the horizon task. The horizon task is an online task in which participants must choose between two slot machines to win the most points possible. One machine has an objectively higher mean of amount of points than the other, unknown to participants. Participants must choose to either explore a slot machine they have less knowledge of points about or exploit the slot machine they know more about. Their choice on the machine they know less about is a measure of directed exploration, but their decision to choose the lesser-known machine is dependent upon the number of trials. Participants can play games with only one free trial or six, known as the horizon. We measured participants aversion to ambiguity by assessing how often they chose the lesser-known machine on horizon 1 and assess their directed exploration by assessing the difference between their choices from horizon 1 to horizon 6. Random exploration is assessed by the number of times participants chose the machine with the lower average mean despite knowing equal amounts of information about the two machines.

Results: We find in our sample that younger adults have better fluid intelligence and older adults have better crystallized intelligence. We also find age group differences in decision making in terms of ambiguity aversion and random exploration. Preliminary results show that neither Crystallized or Fluid intelligence independently mediate the age effect on Random Exploration or Ambiguity Aversion.

Conclusions: Future research is needed to assess if fluid versus crystallized intelligence scores can mediate the effects of suboptimal decision-making in older adults. If decision-making skills are mediated by fluid intelligence, older adults could be taught new ways to utilize and preserve fluid intelligence to make better decisions, potentially reducing decision-making errors within older age groups.

**PERSISTENT UNIVARIATE INDEX BASED ON AGGREGATED COST OF CYCLES IN BRAIN NETWORK.** Farazi M, Zhan L, Lepore N, Thompson PM, Wang Y. Arizona State University; University of Pittsburgh; Children's Hospital Los Angeles; University of Southern California; Arizona Alzheimer's Consortium.

Background: Curse of arbitrary brain network thresholding has been a challenging part of network analysis in medical image community. Persistent Homology, a topological powerful scheme in topology can be a fruitful tool to address this issue. We defined an aggregation cost based on first Betti number, i.e. number of cycles in a planar graph, to track the changes in between group analysis. Based on this feature, we defined a univariate index which enjoys the monotonically increasing property to discover the dissimilarities among brain networks of Alzheimer's Disease (AD), Early Mild Cognitive Impairment (EMCI), Late Mild Cognitive Impairment (LMCI), and normal control subjects (NC).

Methods: The proposed method includes a multi resolution filtration based on edge weights in a graph. First, we filter the graph from empty node set  $V$  to final complete graph by adding the edges increasingly. In this way, the number of connected components and cycles change over the filtration values. We not only localize the cycles using shortest path algorithm but also aggregate the weights of each cycles to define our aggregated cost of cycles (ACC). Finally, we normalize the ACC and multiply it by the number of cycles at each filtration based on Euler characteristics of a planar graph. We applied our method to both synthetic dataset for a better understanding of weakness and strengths of our algorithm and drawing comparisons with other methods in the literature. As for real world application, we applied our method to ADNI dataset of Diffusion Imaging Tractography (DTI) brain networks of 200 subjects.

Results: The univariate index we used is the slope of ACC plot (sACC) and performed permutation test as for statistical analysis of between group analysis. The results based on different and same number of modules created in synthetic dataset show outperformance with respect to previous methods. However, in case of ADNI dataset, the results do not pass the significance level of p-value 0.05 in EMCI/LMCI versus normal subjects which is the case for previous methods as well. As for AD versus NC and MCIs, the results are promising.

Conclusions: Univariate index in brain network based on persistent homology can circumvent the curse of arbitrary thresholding in brain network analysis. In this study we proposed a persistent feature for statistically comparing brain networks in ADNI dataset that shows promising results in discrimination based on AD group versus other groups including NC and MCIs.

**THE DEVELOPMENT OF A DYR533, A HIGHLY SELECTIVE AND ORALLY BIOAVAILABLE INHIBITOR OF DYRK1A FOR TREATMENT OF NEURODEGENERATIVE DISEASE.** Foley C, Dunckley T, Hulme C. University of Arizona; Arizona State University; Arizona Alzheimer's Consortium.

Background: Targeting Alzheimer's disease (AD) pathology at single components is clearly impossible, and a successful therapeutic strategy will require pleiotropic interventions. Herein, we articulate an alternative strategy involving targeting of both amyloid and tauopathies through selective inhibition of the dual specificity tyrosine phosphorylation regulated kinase-1A (DYRK1A), thereby reducing both APP and tau phosphorylation events. The hyperphosphorylation of the microtubule stabilizing protein tau contributes to tau dysfunction and aggregation into NFTs, which are highly correlated to dementia severity in various neurodegenerative diseases such as Alzheimer's Disease.

Methods: Medicinal Chemistry

Results: DYR533, a highly selective and orally bioavailable inhibitor of DYRK1A has been developed.

Conclusions: DYRK1A inhibition by DYR533 offers a promising approach toward the treatment of neurodegenerative disease.

**THE IMPACT OF ALZHEIMER'S FAMILY HISTORY, SEX, AND APOE-4 STATUS, ON WHITE MATTER INTEGRITY IN HEALTHY OLDER ADULTS.** Gallegos N, StickeL A, Ryan L. University of Arizona; University of California, San Diego; Arizona Alzheimer's Consortium.

Background: It is well-documented that white matter integrity becomes compromised with increasing age. In individuals with Alzheimer's disease (AD), certain white matter tracts, including the uncinate fasciculus and the inferior longitudinal fasciculus, appear to decline at a faster rate compared to normal aging, and these tracts have been associated with reduced cognitive abilities including poorer memory.

Methods: The present study investigated how several risk factors for AD – family history, the apolipoprotein e4 allele (APOE-4), and sex – impact white matter integrity in 54 cognitively healthy older adults with and without a family history of AD. White matter integrity was assessed with high angular resolution diffusion tensor imaging, measuring fractional anisotropy, axial, radial, and mean diffusivity. Two white matter tracts, the uncinate fasciculus and the inferior longitudinal fasciculus, were measured based on prior research suggesting that these tracts are affected early in the progression of AD, as well as among mild cognitively impaired individuals.

Results: Using multiple regression models controlling for age and years of education, males and those with the e4 allele had significantly poorer diffusion measures in the inferior longitudinal fasciculus. Additionally, the interaction between family history and sex predicted radial and mean diffusivity in the right inferior longitudinal fasciculus, such that family history only showed a negative effect on diffusivity among males but not females.

Conclusions: These results suggest that risk factors for AD may have differential effects across white matter tracts, and that sex is an important demographic variable to consider in understanding risk for age-related changes in white matter integrity.

**LABEL-FREE ULTRASENSITIVE DETECTION OF AMYLOID-B USING MICROTOROID RESONATORS WITH LIPID SURFACE FUNCTIONALIZATION.** Gin A, Nguyen PD, Li C, Su J.  
University of Arizona; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease is the most common form of dementia with patients exhibiting progressive memory loss and deteriorating mental function. The most frequent forms of Amyloid- $\beta$  peptide, A $\beta$  1-42 and 1-40, aggregate to form oligomers and plaques in the brain, disrupting cell function and ultimately causing cell death. AD patients and non-AD patients have been shown to have differing levels of A $\beta$  in both cerebrospinal fluid (CSF) and serum several years before AD symptoms are displayed, indicating A $\beta$ 's potential as an early predictor for AD. The current gold standard for measuring A $\beta$  levels in CSF and serum has been the ELISA immunoassay but results have had high variability between labs. Therefore, a cost-effective, quick, and consistent assay is desired for detection of A $\beta$  in CSF and serum. In this work, we fabricated microtoroid optical resonators for biosensing because of their high sensitivity and potential for high throughput screening. Each microtoroid sensor is coated with the appropriate A $\beta$  antibody, and A $\beta$  adsorbing onto the sensor surface is detected through its interaction with the evanescent electric field in real time, resulting in a label-free sensor.

Methods: Microtoroid biosensors are first fabricated from SiO<sub>2</sub> on silicon wafers using standard photolithography techniques and CO<sub>2</sub> laser reflow to produce a smooth surface finish -- ideal for low loss optical resonators. Resonance frequencies for each microtoroid are found by evanescently coupling a tapered optical fiber with the cavity and modulating the frequency of a tunable laser (765nm-780nm) at the input of the tapered fiber. The resonance frequencies can be observed on a photodetector as a sharp dip in the transmission and can be tracked using top-of-fringe locking techniques. To prepare the microtoroid for biosensing, the surface of the microtoroid is coated with a lipid bilayer by lipid vesicle fusion. The lipid bilayer provides a smooth, uniform surface which blocks non-specific binding to the surface of the cavity. After the lipid coating, A $\beta$ 42 antibody is covalently bound onto the lipid surface and the microtoroid is placed into a custom constructed wet chamber for biosensing. Concentrations of A $\beta$ 42 in CSF and serum acquired from AD patients are measured by tracking the resonant wavelength shift over time as the solution flows over the sensor surface.

Results: The quality factor (Q-factor) of the microtoroid resonator was measured using the full width at half max linewidth of the transmission dip. Average Q-factor of bare silica microtoroids before functionalization was measured to be 106-107. The increase in surface thickness and surface local refractive index change as a result of bio-functionalization was confirmed by actively tracking the resonance frequency shift towards longer wavelengths and fluorescent imaging. The capability of the biosensor for A $\beta$ 42 detection is confirmed by flowing different concentrations of A $\beta$  1-42 standard solutions over a lipid-functionalized microtoroid, with a limit of detection of 100 aM. The lipid-functionalized microtoroid detected A $\beta$ 42 concentration levels in AD patient CSF at comparable levels to A $\beta$ 42 ELISA assays of CSF samples.

Conclusions: Lipid membrane coated microtoroid optical resonators are shown to be highly capable at detecting low levels of A $\beta$ 42 in AD patient CSF and serum with higher sensitivity and specificity than current biomarker assays for AD. The lipid membrane surface enhances sensitivity by decreasing non-specific binding and allows bio-detection in complex media (i.e. serum).

**CAREPRO-LTC – AN INITIAL PILOT PROJECT.** Gómez-Morales A, Carll P, Goldman J, Gonzalez MS, Coon DW. Arizona State University; Arizona Alzheimer's Consortium.

Background: In 2019, 5.8M people were diagnosed with Alzheimer's and the incidence is growing every year. As the person needs more assistance, people with Alzheimer's disease and related dementias (ADRD) can be placed in assisted living communities making up to 50% of the residents, with 61% of these residents having moderate or severe cognitive impairment. Placing a loved one in a facility does not mean that the caregiving role stops and its stress and distress disappear. CarePRO-LTC addresses the ongoing needs for family and friends caring for individuals with dementia (Alzheimer's disease and related dementias – ADRD) who have been placed in long-term care facilities.

Methods: CarePRO-LTC is an adaptation of the recognized evidence-based caregiver intervention of Care Partners Reaching Out (CarePRO). CarePRO-LTC is a skill-focused program conducted over approximately 5 weeks. It consists of 5 weekly group sessions lasting 2.5 hours each intermingled with 5 weekly individual telephone coach sessions lasting approximately 30 minutes each. For this small pilot, an interactive intervention group was held at a community area in a long-term care facility. Due to differences in facilities and the unpredictable nature of dementia, caregiver participants had the opportunity to learn how to apply the skills offered to different types of stressors by participating in modeling, role-playing, and feedback/debriefing during the group sessions and through skill-reinforcement and feedback during the weekly telephone coach calls. Outcomes investigated were gathered through individual telephone assessments with eligible participants identified through the screening process. These assessments lasted approximately 45 minutes and were administered twice: once after the screening process and within two weeks of the first workshop and within 2 weeks after the end of the intervention (the last coach call).

Results: The results of this small pilot workshop revealed that the participants benefited a great deal from the program (75%), and it helped them to better understand memory loss (100%). Participants found the project helped them to gain confidence (100%) and enhanced their ability to care for their loved ones (100%). As a result, the participants perceived that the workshop improved their loved ones substantially (50%). Last, participating in the program helped to make their lives easier (100%). Post-intervention assessments also revealed the benefits of participating in the pilot workshop such as improving the relationship with their loved one, enriching their experience of being a caregiver, improving communication skills, realizing the need to discuss and plan end of life decisions, and helping them to accept that one cannot be a perfect caregiver.

Conclusions: Preliminary results from the small pilot study suggest high acceptance and satisfaction with the workshop. The intervention was useful for the caregivers and helped them to think about their current situation and future concerns. During the intervention, we learned that caregivers believe they could benefit from more in-depth conversations about end of life decisions.

**NPC DEFECTS IN ALZHEIMER'S DISEASE: INVESTIGATING NUP153, -93, AND -214 MISLOCALIZATION IN HIPPOCAMPAL NEURONS.** Goras M, Brokaw D, Nolz J, Delvaux E, Mastroeni D, Coleman PD. Arizona State University; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease (AD) is a progressive, neurocognitive disorder characterized by memory dysfunction and the presence of senile (A $\beta$ ) plaque and neurofibrillary tangles (composed of phosphorylated tau) [1]. Even though research efforts have contributed insights into the biology of AD, the underlying pathways mediating the cognitive decline are still not completely understood. Eukaryotic cells rely on the movement of proteins between the nucleus and cytoplasm which occurs through the nuclear pore complex (NPC). Nuclear pore complexes (NPC) contain approximately 30 different types of nuclear pore proteins known as nucleoporins (NUPs) that serve as selective barriers between the nucleus and cytoplasm [2]. Mutations in nucleoporin genes have been linked to various human diseases including neurological, nephrotic, cardiac, and neurodegenerative diseases [4]. A recent study showed miss-localization of nucleoporins in AD with direct phosphorylated-tau interaction [8]. Particularly, the report found mislocalization of NUP98 and NUP62 to the cytoplasm, often colocalizing with phosphorylated-tau and suggesting the depletion of NUPs from the nuclear envelope and potentially the direct interaction of phospho-tau [7]. These changes are consistent with NPC deterioration and nuclear-cytoplasmic transport defects [7]. However, this study only investigated four nucleoporins is ill-equipped to make conclusions regarding global changes to the NPC in AD that contribute to AD nuclear dysfunction to pathophysiology. Understanding how NPC subunits, specifically nucleoporins 153, 214, and 93 are affected in AD will expand our understanding of AD cell function and inform therapeutic advances.

Methods: Immunohistochemistry was performed on glycol-fixed 40-micron sections of brain tissue and stained with DAB. Imaging of the CA1 region of the hippocampus at 40x magnification was used to find the optical density of neuron cells using opensource image analysis software ImageJ.

Results: Increased presence of NUP214 in both the nucleus and cytoplasm and presumptive neurotransmitter bundles in AD hippocampal neurons (figure 2). Elevated amount of NUP93 in the nucleus of AD but not ND hippocampal neurons (figure 3). Nuclear localization of NUP153 in AD but not ND (figure 4). Significantly greater optical density of NUP214 and NUP153 in AD compared to ND hippocampal neurons. Unclear whether these NUPs colocalize with phosphorylated tau and promote tau aggregation over the course of the disease (figure 5).

Conclusions: This study showed the mislocalization of cytoplasmic NUP 214 in the cytoplasm, inner ring NUP 93 in nucleus, and suggested aberrant localization of inner basket NUP 153 in the nucleus in AD. A subsequent study is also being implemented to explore NUP intracellular localization in relation to MC1 pathological tau conformation which will inform early diagnostic advances and drug target therapy.

**TREATING MICROGLIA-INDUCED INFLAMMATION WITH MAS RECEPTOR AGONISTS.**  
Hurst CLS, Soto M, Rodgers KE. University of Arizona; Arizona Alzheimer's Consortium.

Background: Neuroinflammation is a common pathology of neurodegenerative diseases, including Alzheimer's Disease, and is believed to be caused by chronically active microglia. Therefore, decreasing inflammation by reducing microglia activation is a promising way to target AD. The Renin-Angiotensin system (RAS) regulates inflammation in the brain through production of free radicals and expression of cytokines and chemokines, and microglia have been found to express all RAS receptors and components. The Mas receptor and its native ligand Angiotensin (1-7), components of the protective axis of the RAS, have anti-inflammatory effects. Here, we characterized the response of microglia in vitro when activated and treated with Mas agonists.

Methods: Using the immortalized microglia cell line HMC3 from ATCC, cells were activated with various stimuli (IFN-gamma, TNF-alpha, PMA/ionomycin, and Angiotensin-II) and treated with various concentrations of the small molecule Mas agonist RASRx1902. Changes in microglia activation were measured with several antibodies, including CellRox, CD68, and HLA-DR.

Results: HMC3 cells showed significant and consistent activation upon addition of IFN-gamma for 48 hours, but not with TNF-alpha, PMA/ionomycin, or Angiotensin-II. Activation was significantly reduced when cells were treated with RASRx1902.

Conclusions: The Mas agonist RASRx1902 was able to significantly decrease HMC3 cell activation in vitro, indicating that it might be effective in decreasing neuroinflammation. Future work will be focused on further characterizing the effects of RASRx1902 in a HMC3 cell line overexpressing the Mas receptor.

**ABCC1 OVEREXPRESSION ENHANCES THE ALPHA-SECRETASE CLEAVAGE OF APP IN VITRO.** Jepsen WM, De Both M, Piras IS, Siniard AL, Henderson-Smith A, Ramsey K, Huentelman MJ. Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

Background: ATP-Binding Cassette protein subfamily C, member 1 (ABCC1), also known as MRP1 (Multidrug Resistance Associated Protein 1), has been suggested as a drug target in Alzheimer's disease because it has been shown to export amyloid beta (Abeta) from the cerebral spinal fluid to the periphery in mouse models. Clinical trials targeting ABCC1 have attempted to increase the transporter's activity to increase the efflux of Abeta from the brain to stall AD progression. Using cell-based assays, we investigated the ABCC1-mediated export of Abeta from the cytoplasm, the extracellular concentrations of Abeta1-40, Abeta1-42, sAPPalpha, and sAPPbeta, and the transcriptomic changes that result from ABCC1 overexpression.

Methods: ABCC1 was cloned into pSBbi-Pur (Addgene) and transfected into BE(2)-m17 human neuroblastoma cells (ATCC), with empty vector serving as negative control. To confirm Abeta export by ABCC1, cells were incubated with HiLyte fluorescent Abeta1-42 (Anaspec), then subject to FACS to count the percentage of fluorescent cells. To investigate the metabolic and transcriptomic changes associated with ABCC1 overexpression, cells were grown in 6-well plates for 4 days when supernatant was then harvested and RNA extracted using the Quick-RNA Miniprep Kit (Zymo Research). All samples stored at -80 °C until 3 separate experiments were performed and assayed together. Abeta1-40, Abeta1-42, sAPPalpha, and sAPPbeta were measured via ELISA (Invitrogen or MyBioSource) or electrochemiluminescence (Meso Scale Discovery). The entire process was repeated four times. Transcriptomic changes were confirmed in ReNcell VM neuroprogenitor cells (Millipore) via TaqMan (Applied Biosystems).

Results: From the FACS experiment, the ABCC1 overexpressing line was 10.7% lower in population fluorescence compared to the control. In all APP metabolism experiments, Abeta1-40, Abeta1-42, and sAPPbeta were significantly decreased (-34.55%, -37.6%, and -25.9%, respectively), while sAPPalpha was only decreased in one experiment. The ratios of sAPPalpha over sAPPbeta indicate that ABCC1 overexpression enhances the alpha-secretase pathway. Transcriptomic analysis identified the significant downregulation of TIMP3 (Tissue Inhibitor of MetalloProteinases 3) as a possible mechanism, as TIMP3 is capable of inhibiting alpha-secretases such as ADAM10 and ADAM17.

Conclusions: ABCC1 is not only capable of exporting Abeta from the cytoplasm of cells, but its overexpression enhances the alpha-secretase mediated metabolism of APP. In all experiments, TIMP3 was significantly downregulated when ABCC1 was overexpressed, and is the likeliest candidate for the mechanism by which ABCC1 modulates APP processing, though the link between ABCC1 and TIMP3 is unknown. Future drug development targeting ABCC1 could work in two ways; first, ABCC1 export activity could be increased to remove Abeta from the brain, and second, modulation of ABCC1 expression (or the mechanism by which ABCC1 alters TIMP3) could increase alpha-secretase activity to prevent further Abeta deposition.

**ASSESSMENT OF ADVANCED NEUROVASCULAR MRI BIOMARKERS IN ALZHEIMER'S DISEASE.** Keeling EG, Bergamino M, Steffes L, Martinez T, Burke A, Baxter L, Su Y, Stokes AM. Barrow Neurological Institute; Arizona State University; Mayo Clinic Arizona; Banner Alzheimer's Institute; Arizona Alzheimer's Consortium.

**Background:** Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by cognitive decline in aging populations. Many magnetic resonance imaging (MRI) techniques have been used to study AD, but these methods are often limited in their sensitivity to underlying neuropathological changes. Recent evidence suggests that neurovascular changes could play a role in the pathogenesis of AD. These changes are thought to occur presymptomatically, as well as prior to other AD-related changes such as atrophy and amyloid pathology. If neurovascular changes are associated with cognitive decline, they could provide novel biomarkers for preclinical assessment of incipient mild cognitive impairment (MCI), a known prodromal phase of AD. The aim of this study is to establish our advanced MRI method for perfusion and functional imaging in cognitively normal and cognitively impaired aging cohorts. This method enables simultaneous acquisition of both micro- and macro-vasculature changes that are thought to play a role in cognitive decline.

**Methods:** Two subject groups were recruited for the study: cognitively normal (CN) (n = 5) and cognitively impaired (CI) (MCI and mild to moderate AD) (n = 4). Subjects underwent cognitive testing using the Montreal Cognitive Assessment (MoCA) and Hopkins Verbal Learning Test (HVLT), along with other cognitive tests prior to MRI. MRI data were acquired at 3T (Ingenia, Philips) at the Barrow Neurological Institute. Structural MRI data was obtained using ADNI (Alzheimer's Disease Neuroimaging Initiative) protocols. Advanced MRI methods were used to measure functional activation (via the fMRI blood oxygen level dependent (BOLD) response) and perfusion metrics cerebral blood flow (CBF) and volume (CBV) (via perfusion MRI acquisition during injection of Gd-based contrast agent). Perfusion MRI and fMRI were acquired using an advanced multi-echo, multi-contrast MRI technique that enables separation and quantification of total and microvascular characteristics. Tasks include vision, face-name encoding and retrieval, and a self-reflection task. Within one month of MRI, a subset of subjects (n = 4) underwent amyloid PET scanning with 18F-Florbetapir at Banner Alzheimer's Institute.

**Results:** Preliminary trends between CN and CI groups are indicative of hypoperfusion in the CI group. These results could be indicative of varying microstructural traits between groups. The control fMRI task (vision) reveals no apparent difference in BOLD response between CN and CI groups, as expected. Analysis of the memory-associated fMRI tasks is ongoing and will be presented at the meeting.

**Conclusions:** This study provides insight into neurovascular changes that may be indicative of AD pathogenesis with the use of a novel multi-scale MR imaging technique. This method provides macro- and microvascular sensitivity to characterize brain perfusion and functional activation. These complementary biomarkers will provide a more comprehensive understanding of neurovascular contributors to Alzheimer's disease and could be potential biomarkers of incipient MCI or AD.

**LONELINESS AND AUTOBIOGRAPHICAL MEMORY SHARING IN OLDER ADULTS.**  
McVeigh KM, Mehl MR, Wank AA, Polsinelli AJ, Moseley S, Glisky EL, Grilli MD. University of Arizona; Indiana University; Minnesota Epilepsy Group; Arizona Alzheimer's Consortium.

Background: Loneliness is conceptualized as a perceived lack of social and emotional support. It is related to many adverse health outcomes, such as depression, anxiety, and cognitive decline, and there is growing interest in the association between loneliness and risk for Alzheimer's disease. Loneliness has not yet been studied in the context of autobiographical memory (i.e., memory for personal events), which has been linked to other AD risk factors (such as older age and APOE status). To begin to bring together these two lines of research, we investigated whether older adults who report being lonelier present with autobiographical memory alterations in daily conversations.

Methods: Participants included 102 cognitively unimpaired older adults (age range = 65-90, M = 76.12, SD = 6.00). We used the Electronically Activated Recorder (EAR) as an unobtrusive, observational method of capturing real-life, everyday instances of memory sharing over the course of four days. Sound files containing conversation were identified and scored for memory sharing using an established protocol. We measured self-reported loneliness with the loneliness scale from the National Institute of Health (NIH) Toolbox. We examined the relationship between loneliness and number of conversations, number of episodic and non-episodic autobiographical memories, and number of details included in episodic memories.

Results: Higher loneliness showed a significant negative relationship to number of conversations ( $r = -.22$ ,  $p = .03$ ) and number of episodic autobiographical memories ( $r = -.20$ ,  $p = .05$ , although this effect was no longer significant if we controlled for number of conversations). However, loneliness was not significantly related to non-episodic autobiographical memory sharing ( $r = -.10$ ,  $p = .35$ ) or number of details included in episodic autobiographical memories ( $r = -.13$ ,  $p = .23$ ).

Conclusions: Initial results found that older adults who report being lonelier had fewer conversations and shared fewer episodic autobiographical memories (albeit not independent of their tendency to have fewer conversations). Future work will explore additional correlates of loneliness in older adults, including the quality of their social interactions and their language use.

**SEMI-AUTOMATED LESION DETECTION SOFTWARE FOR VOLUMETRIC ANALYSIS.**  
Murphy D, Wheeler G, Black J, Bernstein A, Zhou W, Trouard T. University of Arizona; Arizona Alzheimer's Consortium.

Background: Carotid revascularization procedures are effective in stroke prevention, but can lead to the formation and development of microinfarcts in the brain. Conflicting evidence suggest that procedure-related microinfarcts can lead to cognitive decline and that size matters. The typical method for microinfarct quantification involves manually outlining lesions in the brain on diffusion-weighted MRI images and apparent diffusion coefficient (ADC) maps. This procedure can be very time-consuming for radiologists and have low intra- and inter-rater reliability. To address this, we have developed a semi-automatic lesion quantification software capable of automatically determining volumes from point-selected lesions with automatic mapping of lesions onto standard brain atlas.

Methods: This semi-automatic lesion detection software consists of two platforms: MATLAB and FMRIB Software Library (FSL). The user first uploads a DWI and ADC map to a Graphical User Interface (GUI) developed in MATLAB. The user selects a single point within a lesion and the software automatically finds the peak of the lesion and outlines the lesion based off thresholding signal intensity values in the DWI. The software automatically outlines the lesion in three dimensions (in-plane and out of plane). This creates a binary lesion mask that can be used to calculate total lesion volume. Using the subjects T1-weighted image, the lesion mask can be registered to a T1-weighted atlas using FSL. The automatic lesion volumes were compared to those manually outlined in 196 subjects.

Results: Semi-automated lesion volumes had a high showed a high correlation with those manually outlined. Linear regression between the two datasets had an  $R^2 = 0.995$ . Current threshold values in the semi-automated routine calculates lesion volumes that are 19% smaller than those manually determined.

Conclusions: Semi-automatic lesion detection is a highly efficient and accurate approach that has the potential to reduce lesion volume calculation time and increase reproducibility. Future work includes adjusting the lesion volume thresholds to more accurately match manual selection and to conduct timed trials of manual versus semi-automatic lesion circling to quantitatively demonstrate the usefulness of this software for researchers and clinicians.

**THE PREDICTIVE RELATIONSHIP OF SLEEP TO CHANGE IN COGNITIVE STATUS.** Nosker J, Kiefer J, Cornelius A, Zhang N, Auman B, Belden C, Shill HA, Driver-Dunckley E, Mehta SH, Shprecher DR, Sabbagh MN, Beach TG, Adler CH. Banner Sun Health Research Institute; Fielding Graduate University; Mayo Clinic Arizona; Barrow Neurological Institute; Arizona Alzheimer's Consortium.

**Background:** Sleep is considered a modifiable factor of aging and has been a popular area of empirical inquiry. Consequently, sleep is a critical factor to be investigated as it is estimated that 60% to 70% of adults with some form of cognitive impairment have experienced sleep disturbance (Wennberg et al., 2017). Epidemiological research has shown a strong association between insomnia and disorders of aging, including mild cognitive impairment, Alzheimer's disease, Parkinson's disease with dementia, and dementia with Lewy bodies (Pistacchi et al., 2014). Specifically, studies have shown a relationship exists between sleep apnea and Alzheimer's disease (Polsek et al., 2018), daytime sleepiness and vascular dementia (Elwood et al., 2010), and insomnia and all-cause dementia (Chen et al., 2012). Less is known about whether quality and quantity of sleep and daytime sleepiness can predict change to mild cognitive impairment and dementia.

**Methods:** Longitudinal data from the Arizona Study of Aging and Neurodegenerative Disorders and the Brain and Body Donation Program was analyzed to assess the predictive impact of sleep factors prior to the development of cognitive impairment and neurodegenerative disorders of aging. Data from 2009 to 2019 was analyzed to examine the relationship between change in cognitive status and four sleep variables: study participant and partner ratings of daytime sleepiness on the Epworth Sleepiness Scale (ESS), total hours of nighttime sleep (sleep quantity), and total minutes of midnight wakings (sleep quality). Change in sleep variables was analyzed using the two assessments prior to a change in cognitive diagnosis using paired-samples t-tests. Potential covariates of age and time between assessments were examined in relation to change in sleep quality; however, the relationships were nonsignificant and not included in the analysis.

**Results:** Results of the study demonstrated that prior to a change in cognitive status from normal to all cause dementia, a partner's ratings of daytime sleepiness increased significantly ( $M_{change} = 1.60$ ,  $SD = 4.67$ ,  $t(34) = 2.03$ ,  $p = .05$ ). Study participant's ratings of daytime sleepiness, sleep quality, and sleep quantity did not change prior to a change in cognitive status from normal to dementia ( $ps > .05$ ). Partner and study participant's ratings of daytime sleepiness, sleep quality, and sleep quantity also did not change prior to a change in cognitive status to mild cognitive impairment ( $ps > .05$ ).

**Conclusions:** The sequencing of the relationship between sleep and change in cognitive status is a critical factor to be investigated. The implications of this initial study suggest that (a) increased daytime sleepiness may serve as a behavioral marker of cognitive decline, and (b) partner input is an important factor to consider when assessing risk factors for change in cognitive status. Conversely, a patient's self-report of sleep factors may not serve as a reliable behavioral marker of cognitive decline. Future research should include partner ratings of sleep when assessing for predictors of cognitive decline.

**THE IMMEDIATE EARLY GENE EGR3 IS REQUIRED FOR TREM2 EXPRESSION.** Ozols AB, Marballi KK, Beck KL, Morrison HW, Gallitano AL. University of Arizona College of Medicine – Phoenix; University of Arizona; Arizona State University; Syracuse University; Arizona Alzheimer’s Consortium.

Background: Triggering Receptor Expressed on Myeloid Cells 2 (TREM2) is a cell-surface immune receptor on microglia that has recently been linked to Alzheimer’s Disease (AD). Rare coding-region variations in the TREM2 gene increase the risk to develop AD up to 4.7 fold. However, almost nothing is known about the mechanisms that regulate Trem2 gene expression. Early growth response 3 (EGR3) has been identified as a key regulator of differentially expressed genes in AD. Moreover, single-cell transcriptomics of mouse and human brain show that Egr3 gene is highly expressed in microglia. As an activity-dependent transcription factor, the immediate early gene EGR3 is a strong candidate for regulation of the Trem2 gene.

Methods: Analysis of transcription factor binding sites using the TFBind program revealed several high probability EGR3 binding sites in the mouse Trem2 promoter. Electroconvulsive stimulation (ECS), which induces a seizure, was used to activate immediate early gene expression in Egr3-deficient (Egr3<sup>-/-</sup>) and wildtype (WT) littermate mice. An expression microarray study was conducted to identify target genes regulated by Egr3 in the hippocampus. Quantitative real-time PCR (qRT-PCR) was performed to validate the microarray results for the gene Trem2. RNAscope in situ hybridization studies were performed to analyze the expression of Trem2 and Egr3 in the dentate gyrus (DG) and CA1 of the hippocampus in Egr3<sup>-/-</sup> and WT mice following ECS, compared with sham treatment.

Results: The results of our expression microarray study showed that Trem2 was differentially expressed in the hippocampus of Egr3<sup>-/-</sup> mice compared to their WT littermates. Our qRT-PCR studies support the initial finding that expression of Trem2 is deficient in Egr3<sup>-/-</sup> mice compared with WT controls following ECS. RNAscope in situ hybridization studies show that Trem2 and Egr3 co-localize in microglia cells, and levels of Trem2 and Egr3 expression are positively correlated in the hippocampus. Preliminary immunohistologic analyses suggest that Egr3<sup>-/-</sup> mice have reduced levels of hippocampal Trem2 expression compared to WT mice.

Conclusions: Together, these findings suggest the possibility that EGR3, a stimulation-responsive transcription factor, may directly regulate expression of the AD-associated gene Trem2.

**CONTEXT-DEPENDENT MEMORY BETWEEN YOUNGER AND OLDER E4 CARRIERS AND NONCARRIERS.** Palmer JM, Lawrence AV, Bentley Z, Grilli MD, Talboom JS, Huentelman MJ, Ryan L. University of Arizona; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

Background: Distinguishing between highly similar objects requires the use of pattern separation or orthogonalizing information into distinct representations in the brain. Older adults generally perform worse on pattern separation tasks compared to younger adults by incorrectly identifying similar objects as ones seen previously. Therefore, older adults may have a decreased ability to create distinctive representations for objects with many overlapping features compared to younger adults. The background in which an object is placed can lead older adults to make even more similarity judgment errors. Older adults are more likely to make similarity errors when similar objects are embedded in a previously seen background. Our goal was to understand how the apolipoprotein (APOE) e4 allele moderates this effect in both younger and older adults.

Methods: Participants were recruited from an existing pool. Older adults were screened to exclude cognitive impairment. APOE status was determined from saliva via restriction fragment length polymorphism at the Translational Genomics Institute. Objects were embedded in semantically-related scenes and presented one at a time. Participants indicated whether the object was “new,” “similar,” or “same” compared to objects seen previously in the task. Each object was either embedded in a repeated context, a novel context, or on a white background. Behavioral performance was compared between older and younger e4 carriers and noncarriers.

Results: Results indicate that older and younger e4 carriers and noncarriers do not differ on traditional recognition performance. Older and younger adults differed on pattern separation performance such that younger adults were better at identifying similar objects compared to older adults, regardless of e4 status. Participants can either incorrectly identify a similar object as “same” (thinking they have seen this object before; referred to as “Same Error”) or “new” (thinking they have never seen this object before; referred to as “New Error”). Difference scores between these two kinds of errors were calculated, where higher difference scores suggest a tendency to make the “Same Error” more than the “New Error.” We found a main effect of age such that younger adults had lower difference scores than older adults. In addition, there was also a main effect of e4 status such that carriers had lower difference scores than noncarriers.

Conclusions: Taken together, this suggests that the e4 allele may confer a memory benefit that moderates the kinds of errors older and younger adults make.

**AN ASSESSMENT OF THE TRANSITION TO MENOPAUSE IN THE RAT IN THE TGF344-AD MODEL OF ALZHEIMER'S DISEASE.** Peña VL, Northup-Smith S, Woner VE, Bulen HL, Dyer CA, Mayer LP, Bimonte-Nelson HA. Arizona State University; The FYXX Foundation; Arizona Alzheimer's Consortium.

Background: In the United States currently, Alzheimer's Disease (AD) is known to be the sixth leading cause of death with over 5.7 million Americans currently diagnosed. Of those diagnosed, roughly two-thirds are women. It is clear that as our population is aging and AD diagnoses are increasing, we need to further investigate the science driving this increased risk of AD in women. One possible reason for this increased risk is that women undergo a transition to reproductive senescence that results in a drastic decrease in ovarian-derived hormones in contrast to men who undergo a gradual decrease in gonadal hormones over time. Previous preclinical assessments of menopause and AD have been primarily limited to methods of surgical menopause, usually via ovariectomy (the surgical removal of the ovaries). This current project utilized 4-vinylcyclohexene diepoxide (VCD) to model follicular depletion and transitional menopause in the ovary-intact TgF344-AD transgenic rat model of AD expressing two human genes: human amyloid precursor protein (APPSW) and presenilin 1 (PS1E9). This model displays A plaque-like and neurofibrillary tangle-like pathology, neuronal loss. While behavioral assessments have been limited, there is some work showing impairments in transgenic animals compared to wildtype controls. The goal of this experiment was to investigate the relationship between follicular depletion and transitional menopause, and AD-like behavior and pathology.

Methods: Young adult female rats were utilized. Four treatment groups were evaluated: wildtype (WT) VCD (n=9), WT Sham (n=11), transgenic (TG) VCD (n=10), and TG Sham (n=9). Rats received either VCD injections or Sham injections, followed by a behavioral battery that included the Water Radial Arm Maze (WRAM) to assess spatial working and reference memory, the Morris Water Maze (MWM) to assess spatial reference memory, the Open Field Task to assess locomotor activity and anxiety-like behavior, as well as the Visible Platform Task to assess motor and visual acuity to perform a water-escape task. After the behavioral battery, rats were sacrificed and brains, blood serum, uterine horn weight, and ovaries were collected.

Results: WRAM results indicated that TG rats were impaired compared to WT rats during learning, and that TG rats tended to be impaired compared to WT rats during memory retention. TG rats were also impaired compared to WT rats on the MWM. Data are still being analyzed for the Open Field Task. Results for pathological assessments indicated that TG rats with follicle depletion tended to have more beta-amyloid (1-42) in the frontal cortex compared to TG Sham rats.

Conclusions: Collectively, initial data analyses suggest that the TG animals are impaired while learning a complex working and reference memory task in the TgF344-AD model. Further, transitional menopause might influence changes in AD-like pathology in TG rats. Additional analysis of AD-like pathology, ovarian histology, and hormone levels are underway for these behaviorally-tested rats so that relationships between transitional menopause status, behavioral outcomes, and AD-like pathology can be better understood.

**INVESTIGATING SEX-BIASED GENE EXPRESSION IN THE BRAIN TO EXPLAIN ALZHEIMER'S DISEASE MECHANISMS.** Peters M, Natri H, Evanovich A, Buetow K, Wilson M. Arizona State University; Arizona Alzheimer's Consortium.

Background: We are curious if there are any underlying sex differences in the brain that may be related to Alzheimer's disease. Sex differences in gene expression in the brain could be due to genetic differences (genetic females have two X chromosomes while males have one X and one Y), hormonal differences (gonadal hormones such as testosterone, progesterone, and estrogen are notably different between the sexes), and environmental exposure. Here we describe the landscape of sex differences in RNA sequencing data across 11 regions of healthy brains.

Methods: We quantified how different statistical assumptions and the algorithms themselves affected sex-differential expression analysis of RNA seq data derived from 11 types of brain tissue.

Results: Multidimensional scaling indicates sex as the first or second dimension in all tissues. The magnitude and range of differential expression varies by tissue. Overlapping enriched pathways that are upregulated in males (10 out of 11 tissues) are involved in Y-linked functional regulation. For example: regulation of androgen receptor signaling pathway, and histone demethylation. Overlapping enriched pathways that are upregulated in females (5 out of 11 tissues) are involved in dosage compensation. Most enriched pathways that show sex differences are brain-region specific.

Conclusions: There are substantial variations in the magnitude of sex differences across the brain, regardless of which methods are implemented. We aim to next systematically evaluate potential confounders, including; age, pathology, ancestry, and additional phenotypic data. Finally, we will compare differentially expressed genes with Alzheimer's disease genes.

**QUANTIFICATION OF NEURONAL AND ASTROCYTIC CELLS IN THE LOCUS COERULEUS OF COGNITIVELY ASSESSED, YOUNG AND AGED NONHUMAN PRIMATES.** Pyon W, Gray DT, Schnapp W, Schwyhart R, Khattab S, De La Pena NM, Barnes CA. University of Arizona; Arizona Alzheimer's Consortium.

Background: The locus coeruleus is a small brainstem nucleus that is known for its role in supplying noradrenaline to various regions in the brain. The locus coeruleus is especially susceptible to age-related neurodegeneration and is one of the first regions to display Alzheimer's and Parkinson's pathologies (Mather and Harley, 2016), in part due to its high bioenergetic need. Whether differences in number of tyrosine hydroxylase (TH)-expressing neurons significantly contribute to age-related cognitive decline remains less clear.

Methods: To investigate this, coronal brainstem sections from cognitively assessed rhesus macaques (N = 3 aged, mean 28 years; N = 3 adult, mean 11 years) were immunohistochemically labelled to visualize neuronal nuclei (NeuN), catecholaminergic neurons (TH), astrocytes (glial fibrillary acidic protein - GFAP) and vasculature (Solanum tuberosum lectin - STL). For this study, unbiased stereological techniques are used to quantify neuronal numbers. Astrocyte and vascular characteristics are also investigated.

Results: The preliminary results suggest a trend towards lower TH neuron density within the locus coeruleus of aged monkeys. There was also a trend for a relationship between higher TH density and better object recognition memory (delayed nonmatching-to-sample) performance. This trend was not observed with performance on spatial short-term memory (delayed response) or object discrimination tasks.

Conclusions: We are currently examining whether the volume of vasculature within the sampled region, or if properties of astrocytes within the region differ with age. Additionally, we are expanding the number of animals, assessing other neuronal cell types, and evaluating stereological estimates of volume to more thoroughly characterize age-related changes in the locus coeruleus.

**THE PERIPHERAL IMMUNE RESPONSE AFTER A SYSTEMIC INFLAMMATORY CHALLENGE IS NOT INFLUENCED BY ESTROUS CYCLE IN FEMALE MICE.** Rojas Valencia LM, Giordano KR, Tallent BR, Saber M, Jonathan Lifshitz J. University of Arizona College of Medicine-Phoenix; Barrow Neurological Institute at Phoenix Children's Hospital; Phoenix Veterans Affairs Health Care System; Arizona Alzheimer's Consortium.

Background: Endogenous sex hormone levels (primarily estrogen and progesterone) fluctuate to make up different phases of the menstrual cycle. The role of estrogen and progesterone extends beyond the reproductive system, and has been shown to play a regulatory role in the immune system, however, studies provide inconsistent results on the beneficial or detrimental effects of sex hormones. The murine estrous cycle resembles the luteal and follicular phases of the menstrual cycle, which creates translational opportunities to better understand the relationship between the estrous cycle and the immune system. The aim of this study was to determine the influence of the estrous cycle on the proinflammatory response to a peripheral inflammatory challenge.

Methods: Female C57BL/6J mice (n=23) were tracked for estrous cycle phase by daily vaginal smears taken at the same time of day for 8 days prior to LPS administration (1.2 mg/kg, i.p). Blood collected at baseline, before LPS injection (submandibular), and at 24 hours post-injection (terminal blood) was analyzed by flow cytometry to quantify myeloid cell populations and by ELISA to quantify peripheral cytokine levels (IL-6, TNF- $\alpha$ , IL-1 $\beta$ ). Vaginal smears (8 day cycling and 24 hours post-LPS) were stained with Hematoxylin to define the stages of estrous cycle based on observed cell types (neutrophils, cornified cells, epithelial cells).

Results: The levels of IL-1 $\beta$ , TNF- $\alpha$ , and IL-6 increased 24 hours after LPS injection, but the estrous cycle phase did not affect levels of circulating pro-inflammatory cytokines after LPS. Circulating leukocytes (CD45+) and monocytes (CD11b+Ly6Chigh) significantly decreased 24 hours after LPS injection while neutrophils (CD11b+Ly6G+) significantly increased. Estrous cycle phase did not affect levels of leukocytes, monocytes, or neutrophils.

Conclusions: Based on the levels of circulating pro-inflammatory cytokines, leukocytes, monocytes and neutrophils, we could conclude that the peripheral immune response after a systemic inflammatory challenge is not influenced by estrous cycle in female mice

**AGE-RELATED REGIONAL NETWORK PATTERN OF CORTICAL THICKNESS IN HEALTHY MIDDLE-AGED TO OLDER ADULTS.** Smith SG, Bharadwaj PK, Hishaw GA, Trouard TP, Alexander GE. University of Arizona; Arizona Alzheimer's Consortium.

Background: Healthy aging differentially affects cortical thickness, with frontal, temporal and parietal brain regions preferentially affected. Previous studies have focused on univariate analyses to evaluate regional thickness reductions in healthy cohorts over a wide age range, including young to older adults. We sought to use a multivariate statistical method, the scaled subprofile model (SSM; Alexander & Moeller, 1994), to identify a regional pattern of covariance in cortical thickness related to age in a healthy sample of middle-aged to older adults.

Methods: Healthy adults (N = 175; Age = 69.4 + 10.2 years) ages 50 – 89, underwent neuropsychological testing and magnetic resonance imaging (MRI). Cortical thickness for regions of interest (ROI's) were computed in Freesurfer (v5.3) from 3T volumetric MRI scans. Regional SSM network analysis was performed on ROI's using Akaike Information Criteria with 1,500 bootstrap iterations to identify the linear combination of cortical thickness patterns associated with age.

Results: A linear combination of the first eight SSM components was associated with increasing age ( $R^2 = .392$ ,  $p < 1.19e-20$ ). The age-related SSM pattern was characterized by decreases in L inf parietal, L lat occipital, bilat lingual, L parahippocampal, L paracentral, L precuneus, bilat sup temp, and R precentral areas, as well as relative increases in L caudal mid frontal, bilat lat orbitofrontal, L med orbitofrontal, L rostral ant cingulate, L rostral mid frontal, R caudal ant cingulate, and R inf temp areas. After we controlled for age, sex, years of education, and hypertension status, greater expression of the SSM pattern was associated with lower WAIS Full-Scale IQ ( $p = .026$ ), Verbal Comprehension Index ( $p = .021$ ), and Working Memory Index ( $p = .001$ ) scores.

Conclusions: These results indicate a regional pattern of cortical thickness in the healthy community-dwelling older adults with preferential decreases in parietal and temporal brain regions with increasing age, as well as preferential increases in frontal regions. The associations with lower cognitive performance, even after controlling for age, suggests this regional pattern of cortical thickness may influence cognitive aging. That the pattern shows areas with relative increases in frontal cortical thickness may reflect enhanced brain structure in the frontal cortex for those older adults who remain cognitively unimpaired into the eighth and ninth decades of life.

**INTERACTION OF WMH VOLUME AND SEX DIFFERENCES ON HEART RATE RESPONSE TO AEROBIC EXERCISE IN HEALTHY MIDDLE-AGED TO OLDER ADULTS.** Song H, Raichlen DA, Klimentidis YC, Bharadwaj PK, Alexander GE. University of Arizona; University of Southern California; Arizona Alzheimer's Consortium.

Background: White matter hyperintensities (WMH) on magnetic resonance imaging (MRI) scans are associated with brain aging and cerebrovascular disease (CVD), and have been suggested to occur more in women than men. Engagement in aerobic physical activity (PA) may help to slow the development of WMH; however, the extent to which PA influences WMH volume may vary depending on responsiveness to exercise. Here we sought to investigate a novel risk factor for CVD and brain aging by examining whether sex-specific vulnerability to WMH is associated with differences in HR response during exercise.

Methods: Data from the UK Biobank for 813 healthy adults (56.5% women), ages 44-74 years, who completed a submaximal graded exercise test (sGXT) and brain MRI scans and were free of hypertension, diabetes, current smoking, and obesity were included. To determine HR response to exercise, we quantified the change in HR (peak - rest) during the sGXT. WMH volumes were obtained from the UK Biobank after processing with FSL-BIANCA software and were adjusted for intracranial volume. Participants with high vs. low WMH volumes were defined as those above and below the median.

Results: Analysis of covariance showed a significant WMH group by sex interaction for HR response after adjusting for age, BMI, blood pressure, smoking history, and time interval between the sGXT and MRI scans,  $F(1, 803) = 4.57, p = .033$ . The interaction effect remained significant after further adjusting for cardiorespiratory fitness and time spent in moderate-to-vigorous PA,  $F(1, 801) = 4.55, p = .033$ . Simple effects analyses revealed that women with high WMH had greater HR responses than those with low WMH ( $p = .022$ ), whereas men showed no difference; and that among individuals with high WMH, women showed greater increases in HR responses than men ( $p < .001$ ).

Conclusions: The results indicate that larger WMH volumes in healthy middle-aged to older women, but not men, are associated with greater increases in HR during exercise. Our findings suggest that greater chronotropic responses to exercise may be an important biomarker in women, potentially leading to a greater risk for brain aging.

**ROLE OF PREFRONTAL-HIPPOCAMPAL INTERACTIONS IN AGE-RELATED DEFICITS IN SPATIAL WORKING MEMORY.** Srivathsa SV, Khattabi SO, Lester AW, Barnes CA. University of Arizona; Arizona Alzheimer's Consortium.

Background: Neural ensembles in hippocampus and mPFC play a crucial role in memory-guided navigation and decision making, a process susceptible to decline with age in mammals. These regions are connected via a unidirectional projection from the ventral hippocampus to the mPFC and damage or inhibition of this circuit leads to impairments in spatial alternation tasks (Wang et al, 2006). Rats with mPFC lesions show an impairment on spatial working memory tasks (Kim et al., 2009). On the other hand, rats with hippocampal lesions are impaired in both the spatial localization and spatial working memory components (Sapiurka et al., 2016).

Methods: One task that was developed to test the interactions between these regions is a continuous spatial alternation task (Frank et al., 2000), which consists of two interleaved components: an “outbound” component (working memory) and an “inbound” component (spatial memory). Behavioral data from young (9-15 mo) and old (23-30 mo) rats tested on this task reveals that aged rats are slower in learning the inbound component and are unable to learn the outbound component (Kapellusch et al., 2018). The outbound component of the task requires coordination between the hippocampus and mPFC, suggesting that these interactions are impaired in aged rats.

Results: Here, we report data from an ongoing experiment studying the age-associated changes in the hippocampal-mPFC circuit that underlie the behavioral decline in spatial alternation in aged rats through simultaneous electrophysiological recordings in the ventral CA1 region of the hippocampus and in the dorsal ACC region of the mPFC. As disruption of hippocampal SWRs during awake rest leads to impairment of working memory performance (Jadhav et al., 2012) and CA1-mPFC synchronization is stronger during awake SWRs and enhanced in early stages of learning (Tang et al., 2017), several predictions of impact of age on these circuits can be offered.

Conclusions: First, there may be a decrease in the synchronization between CA1 and mPFC unit activity during behavior in aged rats compared to young rats. This is likely to show a strong correlation with the age-related impairments in learning the outbound component. Second, since co-occurrence of hippocampal SWRs and spindles in the mPFC during sleep has been implicated in memory consolidation (Maingret et al., 2016), examining this relationship may provide insights into changes in memory consolidation with age. Furthermore, as increased coupling of the hippocampus and mPFC to the theta and gamma bands is correlated with spatial working memory (Jones et al., 2005; Tamura et al., 2017), we will study the disparities in this coupling during the working memory epochs of the task, between the age groups.

**INFORMANT AND PARTICIPANT-REPORT OF FUNCTIONING PREDICTS PERFORMANCE ON OBJECTIVE COGNITIVE SCREENING.** Stanley AM, Mohebbpour I, Auman B, Belden CM, Virden T, O'Connor K, Callan M, Mansoor J, Evans B, Kiefer J, Roye L, Cline C, Moorley N, Nosker J, Atri A. Banner Sun Health Research Institute; Northwestern University; Arizona Alzheimer's Consortium.

Background: The Montreal Cognitive Assessment (MoCA) (an objective cognitive screening measure) and the Alzheimer's Disease 8 questionnaire (AD8) (a subjective assessment of functional change) are widely used measures for clinical screening of dementia related disorders in individuals 65 and older. Both measures have been identified as reliable predictors of current and/or future dementia. Past research has shown greater accuracy of informant reports in predicting clinician ratings of functional status. However, previous research on the MoCA and AD8 has been solely focused on participant-report AD8 measures without consideration of informant reports. Moreover, participant-reports can be impacted by anosognosia (i.e., lack of insight), which is often observed in individuals with cognitive impairments and is not thoroughly addressed in previous research. This study aims to assess if informant subjective reports can accurately predict their participants overall cognitive functioning.

Methods: Participants ( $N = 212$ ) were seen from 2018 to 2020 through a free community screening service (Brain Health Check-In) at Banner Sun Health Research Institute in Sun City, AZ. We hypothesize that informant reported AD8 and participant MoCA scores will be inversely related (i.e., AD8 is high, indicating greater functional decline, when MoCA is low, indicating greater cognitive impairment). In contrast, we predict the participant-reported AD8 will be weakly associated with MoCA performance due to potential anosognosia. Moreover, we hypothesize the strength of the relationship between the informant reported AD8 and the MoCA, as well the participant-reported AD8 and the MoCA, will vary based on cognitive performance classification (e.g., CN  $\geq 27$ , Mild 25-26, Moderate 23-24, and Highest  $\leq 23$ ). Furthermore, we hypothesize the informant reported AD8 will be a more reliable predictor of cognitive performance. Analysis of data was conducted using SPSS-21. First and second hypotheses were analyzed with Spearman's Rho ( $r$ ), and third hypothesis was tested through a linear regression.

Results: Both the participant and informant reported AD8 directly correlated with overall cognitive performance classification (i.e., rating of functional decline was low when MoCA score was in low impairment ranges) ( $r = 0.639$  [informant],  $p < .000$ ;  $r = 0.610$  [participant],  $p < .000$ ). Informant reported AD8 ratings were significantly inversely correlated with MoCA performance (i.e., informant AD8 score was high when participant MoCA score is low) ( $r = -0.497$ ,  $p < .000$ ). Participant reported AD8 ratings also inversely correlated with overall MoCA scores, but with a weaker association ( $r = -0.296$ ,  $p < .000$ ). Neither participant nor informant reported AD8 were able to reliably predict categorical cognitive performance classification (e.g. normal cognition, mild impairment). However, the informant reported AD8 ( $r = -.686$ ,  $p < .000$ ) did emerge as a reliable predictor of MoCA performance when all cognitive groups were combined, and within Moderate ( $N = 58$ ) and High ( $N = 112$ ) classification levels of cognitive impairment.

Conclusion: This study extends prior research about AD8 and suggests both informant- and participant-reports are valuable; however, informant often provides more clinically useful information. Additionally, these findings reaffirm previous research that anosognosia is often observed in individuals who are cognitively impaired. However, more research is needed to determine at what level of cognitive functioning that insight begins to potentially diminish; as well as, further research is needed to ascertain the predictability of informant reported AD8 on the categorical cognitive performance levels.

**PARTICIPATION IN A COMPREHENSIVE LIFESTYLE PROGRAM AND THE EFFECTS ON QUALITY OF LIFE AND COGNITIVE FUNCTION IN ELDERLY ADULTS.** Tatem A, Sharifi A.  
University of Arizona College of Medicine-Phoenix; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease (AD) is the sixth leading cause of death in the U.S. There is abundant literature on benefits to the aging brain from lifestyle factors. However, there is no direct evidence to indicate if an approach utilizing six lifestyle components (physical exercise, nutrition, cognitive stimulation, sleep, socialization, and stress reduction) in-concert confers quality of life, physical benefits, and cognitive benefits in an aged population. This type of lifestyle program has not been investigated in a continuing care residential community (CCRC) population. The primary aim of this study was to determine the effect of a comprehensive lifestyle program on perceived quality of life in elderly adults, as compared to a control group receiving standard health advice. The secondary aim was to assess the effect on cognitive function using physical fitness as a surrogate marker.

Methods: Participants were recruited from an independent living facility for this prospective cohort study. The intervention group attended a one-hour lesson focused on six lifestyle factors (physical exercise, nutrition, cognitive stimulation, sleep, socialization, and stress reduction) two times per week for a total of approximately 40 sessions. They were encouraged to pursue objectives of the program outside of class. The control group received standard health advice. The WHO-QOL survey and two physical fitness measurements (Timed Up and Go and 30-Second Chair Stand Test) were obtained at baseline, 3 months, and 6 months. We anticipated 40 subjects to participate. The power analysis showed 34 participants were required to achieve a power of 80%. The Wilcoxon Rank Sum compared continuous variables.

Results: There were 41 subjects enrolled (27 intervention vs 14 control), 40 that completed data collection, and 40 that were analyzed. There was a statistically significant improvement in the physical domain score of the World Health Organization Quality of Life (WHO-QOL) assessment tool from baseline to 6 months. The improvement in the environmental domain score approached statistical significance. There was no statistically significant change in the 30 Second Chair Stand Test. Both the control and intervention groups had statistically significant improvements in the Timed Up and Go (TUG) test.

Conclusions: Participation in a six-component lifestyle program improved quality of life of elderly adults living in a CRCC. Specifically, subjective physical health improved with participation. This study indirectly supports lifestyle factors in the fight against dementia, as improved physical health is correlated with reduced cognitive decline. Inconclusive results in other WHO-QOL domains and physical tests may be due to the small sample size, short study duration, other prior and ongoing physical activities, and/or residing in an enriched environment.

**DIFFEOMORPHIC CORTICAL SURFACE REGISTRATION WITH RETINOTOPIC MAPPING: A PRELIMINARY STUDY FOR QUANTIFYING PRECISE VISUAL CORTICAL MORPHOLOGY CHANGES OF ALZHEIMER'S DISEASE.** Tu Y, Ta D, Gu X, Lu Z, Wang Y, Arizona State University; State University of New York at Stony Brook; Harvard University; NYU Shanghai, New York University; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease (AD), is the most common cause of dementia among older adults. One common method to detect AD is to use magnetic resonance imaging (MRI) based surface analysis, which usually quantifies the entropy of the cortical surface. Usually, the cortical surface-based method needs to register/align cortical surfaces across subjects to detect vulnerable regions to the AD. Although sophisticated methods exist, e.g. Freesurfer, FSL, and Brainsuit, it is very challenging to align the cortical surface very well with the only anatomical MRI. Without a precise alignment, the entropy of mild cognitive impairment is difficult to detect as the small changes of surface entropy. The retinotopic map is a research topic that relates the visual input to the cortical surface. Since the retinotopic maps share a common visual space, it provides a common coordinate for visual cortical regions. Prior research has indicated that cortical alternations under age-related visual deficits induces changes on visual cortex which can be measured by retinotopic mapping. We see there is an opportunity to use retinotopic maps as an imaging biomarker for AD research. Unlike conventional surface registration where only scalar features are available, retinotopic mapping associates an estimated visual coordinate (although noisy) to each location of the visual cortex. Second, the quality of the estimated visual coordinates can be assessed with performance metrics, which can help us emphasize high-quality locations and under-weight poor-quality locations. We adopt the retinotopic maps data to improve the alignment of visual cortical regions. This will eventually help the analysis of the impact of Alzheimer's to visual cortex morphometry.

Methods: We follow the common practice of cortical surface registration, i.e. the diffeomorphic condition: cortical surfaces can be aligned by stretching or/and shrinking but without tearing the cortical surface up. Specifically, the diffeomorphic condition is quantified by the Beltrami coefficient. Then we modeled the registration as an optimization procedure of energy function, consisted of features (e.g. cortical thickness, surface curvature, etc.) similarity, and regularization (include the smooth and diffeomorphic constraints). We provided numerical steps to solve the optimization problem. We tested our registration on a synthetic dataset and surfaces with retinotopic data from the Human Connectome Project. There are mainly two differences between the proposed registration and conventional methods: (1) Conventional methods treat all the feature information, e.g. cortical thickness, as a scalar. Although it is the most common case with wide applications, it has not fully used the common coordinates of retinotopic maps. Instead, we directly utilize the common coordinates of retinotopic maps; (2) Instead of using the determinate of the Jacobian matrix, we use the Beltrami coefficient to monitor the diffeomorphic condition. This provides a quantification of the registration angle distortion.

Results: We compared our method with popular surface/image registration methods for the synthetic data, including TPS (mean error in visual coordinates 4.47, the similar meaning of these numbers for later methods), D-Demos (1.20), LDDMM (0.64), and QCHR (0.10). We found the proposed algorithm achieves the smallest registration error (0.08) and ensures the diffeomorphic condition. We also apply the method on the retinotopic data from the Human Connectome Project retinotopic dataset. We achieved a better result than Freesurfer. Also, based on our method, we further improved a template for the visual cortex.

Conclusions: The proposed registration method can reduce surface registration error while ensuring the diffeomorphic condition with the retinotopic fMRI data. Further, the method can adopt hand-drawn landmarks. It is promising to detect subtle morphology changes in the visual cortex influenced by AD progress.

**TOPOLOGICAL SMOOTHING AND PRECISE QUANTIFICATION FOR HUMAN RETINOTOPIC MAPPING: A PRELIMINARY STUDY FOR THE EARLY DETECTION OF ALZHEIMER'S DISEASE.** Tu Y, Ta D, Lu Z, Wang Y. Arizona State University; NYU Shanghai; New York University; Arizona Alzheimer's Consortium.

Background: Alzheimer's disease (AD), is the most common cause of dementia among older adults. It is commonly believed that treatment could have great benefits if it is started during the earliest stages of AD. Therefore, there is a need to develop sensitive detection methods for evaluating AD burden, progression and response to interventions. Although several approaches have proposed to early detect the AD including magnetic resonance imaging-based analysis, protein component analysis, etc., little is known about the visual defects in the aging and Alzheimer's disease. Recently, there are works show that compare to normal aging, the AD makes visual retinotopic maps organizational deficits. Unfortunately, the retinotopic maps, derived from analyzing fMRI signal to visual stimuli, is of noise and topological violations. The big noise halts the precise analysis of retinotopic maps, as well as the feasibility to detect the AD.

Methods: To generate precise retinotopic maps, we adopt the Beltrami coefficient to quantify the topological condition, and then model the smoothing process as an optimization procedure with topological constraints. We provided numerical steps to solve the optimization problem. We tested our algorithm on a synthetic dataset and Human Connectome Project 7T retinotopy. The synthetic data is generated using an ideal retinotopic mapping model (ground truth) with added noise. With the topological smoothed result, we can quantify the visual retinotopic maps in precise.

Results: We compared our method with popular spatial smoothing methods for the synthetic data, including Average Smoothing, Median Smoothing, and Laplacian Smoothing. The application of our proposed method to the synthetic data generates diffeomorphic results without any big deviations, while other methods violate the condition. We also compared the proposed method with spatial methods. We also generate the topological result, especially near the fovea. Based on the result of our method, we estimated the precise quantification of retinotopic maps, including the cortical magnification factor, angle distortion, etc. Also, we see the feasibility to detect the non-topological regions by analyzing the error after smoothing within a specific patch on the cortical surface.

Conclusions: The proposed topological smoothing method can ensure the topological condition and then detect the defects of visual retinotopic maps. The proposed method shows a promise to early detect the AD by analyzing the visual retinotopic maps

## **THE ROLE OF REGIONAL WHITE MATTER HYPERINTENSITIES AND APOE E4 STATUS IN THE MEDIATION OF AGE AND HIPPOCAMPAL VOLUME IN HEALTHY OLDER ADULTS.**

Van Etten EJ, Bharadwaj PK, Hishaw GA, Trouard TP, Alexander GE. University of Arizona; Arizona Alzheimer's Consortium.

Background: While white matter hyperintensities (WMH) have been associated with hippocampal atrophy, less is known about how the regional distribution of WMH may differentially affect hippocampal volumes in healthy aging. Apolipoprotein E (APOE)  $\epsilon$ 4 carriers may be at an increased risk for WMH and greater hippocampal atrophy. The present study sought to investigate whether regional WMH mediate the relationship between age and hippocampal volume and whether this relationship is moderated by APOE  $\epsilon$ 4 status in healthy aging.

Methods: A cohort of healthy adults ( $n=192$ , 94F/98M, mean $\pm$ sd age= $70.5\pm 10.1$ , mean $\pm$ sd Mini-Mental State Exam= $29\pm 1.2$ , APOE  $\epsilon$ 4 status (yes/no) = 59/133), 50 to 89 years of age were included. T1-weighted 3T volumetric MRIs were obtained and processed using Freesurfer (v5.3) software to obtain hippocampal volumes averaged across hemispheres. WMH in the four cerebral lobes were computed using T1 and T2 FLAIR scans and a lesion segmentation toolbox (Schmidt et al., 2012) with Statistical Parametric Mapping (SPM12). Total intracranial volume was computed for each participant using SPM12 to adjust hippocampal and WMH volumes for differences in head size. Mediation analyses were conducted with PROCESS macro software (Hayes, 2012) on SPSS, using bootstrap resampling with 10,000 iterations to produce bias corrected 95% confidence intervals.

Results: Temporal lobe WMH significantly mediated the relationship between age and average hippocampal volume, and this effect was moderated by APOE  $\epsilon$ 4 status ( $-.02$  (SE=.01), 95% CI,  $[-.04, -.003]$ ). APOE  $\epsilon$ 4 carriers, but not non-carriers, showed negative indirect effects of age on hippocampal volume through temporal lobe WMH (APOE  $\epsilon$ 4 carrier:  $-.02$  (SE=.01), 95% CI,  $[-.03, -.003]$ ; APOE  $\epsilon$ 4 non-carrier:  $.00$  (SE=.01), 95% CI,  $[-.01, .02]$ ). These findings remained significant after additionally adjusting for sex, years of education, and hypertension status. There were no significant mediation effects for frontal, parietal, and occipital lobe WMH, with or without covariates.

Conclusions: The results indicate that the effects of aging on hippocampal volume are mediated by WMH regionally localized to the temporal lobes and that this effect depends on APOE  $\epsilon$ 4 carrier status. Together, these findings suggest that differences in hippocampal volumes observed in the context of healthy aging may be in part related to the influence of APOE  $\epsilon$ 4 on WMH and associated vascular mechanisms. Further research is needed to evaluate how regional WMH may influence other neuroanatomical effects of brain and cognitive aging.

**WHITE MATTER INTEGRITY MEDIATES THE RELATIONSHIP BETWEEN BODY MASS INDEX AND EXECUTIVE FUNCTION IN HEALTHY OLDER ADULTS.** Van Etten EJ, Bharadwaj PK, Hishaw GA, Trouard TP, Alexander GE. University of Arizona; Alzheimer's Consortium.

Background: High body mass index (BMI) is a known risk factor for cerebrovascular disease. Elevated BMI has been correlated with brain and cognitive aging, including reductions in white matter integrity (WMI) and in executive cognitive abilities. However, relations between BMI and executive function have not been consistently observed. We sought to further investigate this association by testing whether WMI, as measured by average fractional anisotropy (FA), has a mediating role between BMI and the executive functions of working memory, switching and inhibition in healthy older adults.

Methods: A cohort of healthy adults (n=195, 100F/95M, mean±sd age=69.8±10.6, mean±sd Mini-Mental State Exam=29±1.2, mean±sd BMI 25.4±4.0), 50 to 89 years of age were included. Volumetric T1 and diffusion weighted 3T MRI scans were processed using Freesurfer (v5.3) and TRACULA (Tracts Constrained by Underlying Anatomy; Yendiki et al, 2011) to compute an average estimate of FA from all 18 major white matter tracts. White matter hyperintensity (WMH) volumes were measured using T1 and T2 FLAIR scans and the lesion segmentation toolbox (Schmidt et al., 2012). Mediation analyses were performed with PROCESS macro software (Hayes, 2012) in SPSS with age, sex, and education as initial covariates. Vascular risk factors including hypertension, smoking, and cholesterol status, and total WMH volume were subsequently added as covariates.

Results: There were no direct effects of BMI on executive function. However, FA significantly mediated the relation between BMI and WAIS working memory index (-.09(SE=.06), 95% CI, [-.228, -.002]) and Trail Making Test B (.29(SE=.17), 95% CI, [.012, .680], but not Stroop Word-Color Interference. All findings remained significant after additionally adjusting for vascular risk factors.

Conclusions: Although there were no direct effects, FA mediated associations for working memory and switching, but not inhibition measures of executive function in the relation with BMI. These findings suggest BMI has indirect effects on selective executive functions through its impacts on WMI, supporting BMI as an important factor influencing brain and cognitive aging.

**EVERYDAY AUTOBIOGRAPHICAL MEMORY SHARING: A NATURALISTIC OBSERVATION STUDY OF OLDER ADULTS' DAILY CONVERSATIONS.** Wank AA, Mehl MR, Andrews-Hanna JR, Polsinelli AJ, Moseley S, Glisky EL, Grilli MD. University of Arizona; Indiana University School of Medicine; Minnesota Epilepsy Group; Arizona Alzheimer's Consortium.

Background: Autobiographical memory retrieval has critical implications for everyday social interactions and serves a variety of adaptive functions in daily life. Prior research conducted in the laboratory has found that older adults demonstrate lower episodic specificity, both through the retrieval of fewer autobiographical episodic memories and a reduction in episodic details during elaboration of these memories. Considering that much of this research has been done in the laboratory, relatively little is known about age-related reductions in episodic specificity in the context of daily social interactions. The goal of the present study was to examine daily autobiographical memory and future thought sharing in older adults using a naturalistic observation approach.

Methods: We used a smartphone application known as the Electronically Activated Recorder, or "EAR", to unobtrusively capture real-world conversations over four days. In a sample of 102 cognitively normal older adults (age 65 to 90), we identified instances where episodic or semantic memories and/or future thoughts were shared by participants and scored the episodic memories or future thoughts for episodic and semantic details.

Results: Results revealed that older age was associated with decreased real-world sharing of both autobiographical episodic and semantic memories, independent of the total number of conversations with others. We also found that older age was linked to less detailed elaborations of autobiographical episodic memories. Frequency and detailed descriptions of shared future thoughts did not yield any relationships with age. Similar to prior laboratory research, other analyses showed a possible connection between episodic details shared in autobiographical episodic memories and future episodic thoughts, and no correlation between autobiographical episodic detail sharing and a clinical episodic memory assessment. In contrast to laboratory studies, episodic detail production while sharing autobiographical episodic memories was unrelated to working memory and did not demonstrate gender differences.

Conclusions: Within an older adult sample, our findings provide novel evidence that there may be an age-related decrease in the frequency and detail of autobiographical memory sharing when assessed unobtrusively and objectively "in the wild".

**THE COGNITIVE EFFECTS OF THE HIGHLY SELECTIVE PROGESTIN SEGESTERONE ACETATE IN A RAT MODEL OF SURGICAL MENOPAUSE.** Woner VE, Koebele SV, Northup-Smith SN, Willeman MN, Barker C, Schatzki-Lumpkin A, Valenzuela Sanchez M, Bimonte-Nelson HA. Arizona State University; Translational Genomics Research Institute; Arizona Alzheimer's Consortium.

Background: Progestins, synthetic hormones that mimic the effects of endogenous progesterone, are prescribed for a myriad of health reasons to women across their reproductive and menopausal years. While our laboratory has demonstrated spatial memory deficits following chronic administration of some progestogens, which include progesterone and synthetic progestins, other progestogens have been shown to have neutral or even beneficial cognitive effects. Other laboratories have also reported variable cognitive outcomes, dependent upon variations in experimental design and animal models used; of note, the potential mechanisms behind these differing cognitive outcomes are unknown. The various parent molecules from which progestins are derived are known to confer unique profiles of biological activities and affinities for each progestin beyond the progesterone receptor (PR), and could modulate cognitive outcomes. These findings, along with research demonstrating the role of some progestogens with strong affinities for the PR in neurogenesis and neuroprotection, suggest that a 'purely-progestational' molecule that maximizes PR affinity and minimizes affinities to other receptors may be cognitively beneficial.

Methods: We evaluated the cognitive effects of segesterone acetate (registered trade name: Nestorone, NES; ST-1435), a 19-norprogesterone derivative with a strong PR affinity and no androgenic or estrogenic receptor affinity, in a rat model of surgical menopause. Middle-aged female Fischer-344 rats were given Sham surgery or Ovariectomy (Ovx) followed by daily administration of either medroxyprogesterone acetate (MPA; previously shown by our laboratory to induce cognitive deficits), NES, given at a low or high dose, or Vehicle (administered to the Sham group and one Ovx group). Rats were tested on a behavioral battery that evaluated spatial working and reference memory performance, including the Water-Radial Arm Maze (WRAM), Morris Maze (MM), Visible Platform control task, and Open Field Test to measure locomotor activity and anxiety-like behaviors. At the conclusion of behavioral testing, animals were sacrificed, and tissues from the Dorsal Hippocampus were processed for relative expression of insulin-like growth factor 1 receptor (IGF-1R) via Western Blot.

Results: WRAM results indicated that Ovx rats given the low dose of NES demonstrated impaired spatial working memory compared to Ovx rats given vehicle. Ovx rats treated with the high dose of NES exhibited delayed memory retention deficits on the WRAM, as did MPA-treated Ovx rats, replicating previous findings. On the MM, Ovx rats given MPA or the high dose NES demonstrated impaired reference memory relative to Ovx rats given vehicle. Additionally, unique correlations between Western Blot results and WRAM performance were found where greater IGF-1R expression in the Dorsal Hippocampus correlated with improved WRAM performance during learning for Sham animals.

Conclusions: The cognitive effects and underlying neurobiological mechanisms of NES merit further study, as the memory deficits reported here at both low and high doses contrast with other literature suggesting neuroprotective effects of NES. Thus, we seek to characterize the relationship between the biological activity of NES and subsequent behavioral outcomes in future studies in order to identify a progestin that promotes positive health outcomes for women.

**ASSESSING AMYLOID BURDEN WITH HIPPOCAMPAL MORPHOMETRY FROM VOLUMETRIC MRI DATA.** Wu J, Gui J, Su Y, Chen K, Ye J, Caselli RJ, Reiman EM, Wang Y. Arizona State University; Banner Alzheimer's Institute; Mayo Clinic Arizona; University of Michigan; Arizona Alzheimer's Consortium.

Background: The accumulation of beta-amyloid plaques (A $\beta$ ) in human brains is one of the important hallmarks of AD and this accumulation might be a reason for the structural damages on some subcortical areas, such as hippocampus. Therefore, the morphometry information from volumetric magnetic resonance imaging could be a measure of A-beta burden in the brain. In this study, we studied the feasibility that whether our hippocampal morphometry generated by volumetric magnetic resonance could be a potential biomarker to assess A-beta burden.

Methods: With the hippocampal structure from structural MR images, we calculated our morphometry features, multivariate tensor-based sub-cortical morphometry statistics (MMS), from each hippocampal surface. MMS consists of radial distance (RD) and surface multivariate tensor-based morphometry (mTBM), which takes advantage of the vertex-wise changes in both hippocampal thickness and regional surfaces. With these features, we designed two different kinds of experiments for validation. We firstly performed a Hotelling's T2 test and find a significant difference between the two groups of each cohort. Meanwhile, by applying a correntropy-based sparse coding system, we generated a representation for each subject and achieved superior performance in binary classification of all the four cohorts.

Results: The data used in this paper were downloaded from the Alzheimer's Disease Neuroimaging Initiative (ADNI) project and Open Access Series of Imaging Studies (OASIS). The A $\beta$  positivity was determined using mean-cortical SUVR (standard uptake value ratio) with the cerebellum as reference region. Then, we combined these groups of subjects into four cohorts as AD+ vs. NL-, MCI+ vs. MCI-, NL+ vs. NL- from ANDI, and NL+ vs. NL- from OASIS. All the high-resolution MRI scans were acquired from ADNI 1, ADNI GO, ANDI 2 and OASIS. For the group different study, we performed the Hotelling's T2 test on each side of the hippocampus for all the four cohorts. The left hippocampus p-values for the four cohorts, AD+ vs. NL-, MCI+ vs. MCI-, NL+ vs. NL- from ANDI, and NL+ vs. NL- from OASIS are <0.0001, 0.0476, 0.0492 and 0.0468. And the right ones are <0.0001, 0.0035, 0.1963 and 0.2034. For the second method, we generate one representation for each subject by using the features from both sides of hippocampus. And then, we employed a binary classification on the two groups of each cohort with random forest and employed 10-fold cross-validation to evaluate the accuracy. The results of F1 score for the four cohorts are 0.92, 0.89, 0.74 and 0.84.

Conclusions: Our results demonstrate that our surface multivariate morphometry statistics (MMS) has a superior performance in both statistical study and machine learning study. It could be applied as a potential biomarker for distinguishing patients with different beta-amyloid plaques status.

**EXPERIMENTAL TBI INDUCES LONG TERM COGNITIVE DEFICITS AND VASCULAR PATHOLOGY.** Young C, Law LM, Karamanova N, Truran S, Quarles C, Migrino RQ, Lifshitz J. University of Arizona; Barrow Neurological Institute; Phoenix Children's Hospital; Phoenix VA Hospital System; Arizona Alzheimer's Consortium.

Background: Traumatic brain injury (TBI) is a worldwide health concern with approximately 5.3 million Americans currently living with TBI-induced disability. For many, TBI-associated disabilities impair cognition, provoke seizures, and elevate risk for neurodegenerative disease. Oxidative stress, nitrative stress, and derangement of cerebrovascular physiology have been implicated in acute acquired neurological injury and aging-related dementia disorders. Experimental models of TBI provide a unique opportunity to investigate relationships between vascular disturbances and impaired cognitive performance.

Methods: In this study, diffuse brain-injured (midline fluid percussion) adult male rats were assessed for long-term molecular, vascular, and cognitive pathologies. At 90 and 180 days post-injury, brain-injured and uninjured sham rats performed three cognitive tasks: a novel object recognition task (NOR) assessing short term memory, a novel location recognition task (NOL) assessing long term memory, and a temporal order recognition task (TOR) assessing working memory. Rats' brains were imaged at 182-187 days post-injury via dynamic susceptibility contrast monocrySTALLINE iron oxide particle (MION) magnetic resonance imaging (MRI) to assess in-vivo cerebrovascular blood flow and vasoreactivity in response to hypercapnic challenge. At 189-193 days post-injury, brains were collected for histology and arteries (Circle of Willis, ventral circuit) were collected for ex-vivo vasoreactivity and markers of oxidative and nitrative stress, respectively.

Results: Brain-injured rats performed significantly worse than shams on the NOR and TOR tasks at 90 days post-injury. At 180 days post-injury, brain-injured rats performed significantly worse than sham on all three cognitive tasks. By MRI, brain-injured rats showed significantly less cerebrovascular blood volume compared to sham rats. Ex-vivo vasoreactivity was not significantly different in baseline cerebrovascular myogenic tone or endothelial reactivity between groups. Cerebral arteries of brain-injured rats exhibited significantly greater levels of superoxide and peroxynitrite in the presence of amyloid-beta-42 (A $\beta$ 42) compared to cerebral arteries of sham rats, indicating significantly higher response to oxidative and nitrative stress, which supports the potential for injury-induced vascular dementia.

Conclusions: Further analysis will integrate the relationship between brain injury parameters, cognitive performance, and vascular function in order to elucidate the mechanistic role of vascular dysfunction in TBI-mediated cognitive dysfunction.

**MEDIN OLIGOMERS FORM MEMBRANE PORES WITH POTENTIAL CONSEQUENCES FOR VASCULAR DYSFUNCTION RELATED TO NEURODEGENERATION.** Younger S, Jang H, Davies HA, Niemiec MJ, Garcia JGN, Nussinov R, Migrino RQ, Madine J, Teran Arce F. University of Arizona; University of Liverpool; Phoenix Veterans Affairs Health Care System; Leidos Biomedical Research, Inc.; Sackler School of Medicine, Tel Aviv University; Arizona Alzheimer's Consortium.

Background: Age is the most important risk factor for neurodegenerative diseases, including cerebrovascular disease (CVD) and vascular dementia (VaD). Medin, one of the most common aging-related amyloidogenic proteins, is a 50 amino acid peptide that accumulates in the vasculature with aging. We recently found vascular dementia patients to have an increased concentration of cerebral arterial medin, implicating medin in cerebrovascular degeneration. Neuroinflammatory responses have also been correlated with medin concentration. We attempted to elucidate what similarities in structure and function medin has to amyloid  $\beta$ , and what role these might play in AD.

Methods: We used lipid bilayer electrophysiology (BLM) to show medin oligomers induce ionic membrane permeability by pore formation. We also imaged medin aggregation states with atomic force microscopy (AFM). We used molecular dynamics (MD) simulations to model the structure of medin pores in silico. Finally, we used Thioflavin T fluorescence measurements to characterize the medin aggregation process.

Results: Our BLM results showed that growth-phase medin induces substantial ion conductive pore activity, with reduced pore formation in plateau-phase and minimal in lag-phase. AFM images showed non-fibrillar, flat domain structures reminiscent of supported lipid bilayers. Thioflavin T fluorescence for medin was substantially lower than amyloid  $\beta$  fibrils, reinforcing the lack of fibrillar structures in medin aggregates and pointing to pores following a non-amyloidogenic pathway. The MD simulations provided us with a detailed CNpNC barrel topology giving a similar diameter to our measured pore conductances.

Conclusions: We demonstrate that medin is capable of ion-conductive pore formation in a similar manner to amyloid  $\beta$ , suggesting potential interactions and similar disease pathologies. Thioflavin fluorescence and AFM both show a distinct lack of amyloid fibers in their aggregates, pointing to a non-amyloidogenic aggregation pathway in pore formation. Thioflavin fluorescence and AFM both show a distinct lack of amyloid fibers in their aggregates, pointing to a non-amyloidogenic aggregation pathway in pore formation.

**A DEEP LEARNING MODEL TO PREDICT DRUG-GENE INTERACTION.** Zeng Y, Prinz J, Wang Y, Wang J. Arizona State University; Mayo Clinic Arizona; Arizona Alzheimer's Consortium.

Background: Drug-target interaction (DTI) is an important step of the drug discovery process, which often involves a large number of time- and cost-intensive biological experiments and tests to identify new drugs or novel targets for existing drugs. However, even with all the effects, effective drugs are not available for some specific disease, like Alzheimer's disease. Computational methods can help screen new drugs, or purpose drugs for different disease. In past decade, much effort already has been devoted to developing the similarity-based and deep learning-based approaches for DTI prediction. In this report, we present a deep learning-based method which takes advantage of information fusion to exploit essential feature of drug-gene pairs for DTI prediction. We believe the model would be applied to discover putative drugs that can target over 600 AD associated genes.

Methods: Deep learning methods have achieved breakthrough success in many fields, such as image processing, natural language processing, which fully reflects its powerful feature extraction and feature classification ability. In this work, we try to apply deep learning methods into drug-gene interaction predictions. Firstly, since drug data and gene data have different feature distributions, two PCAs are used to extract individual features in their latent spaces, and also reduce dimensionality. Then, based on the globality of Fully Connected networks (FNN) and locality of Convolutional Networks (CNN), we employ the FNN and CNN blocks to learn individual features from PCA features at the same time. Following the separated feature extracting layers, there are two information fusion layers. The first information fusion layer is utilized to merge outputs of CNN and FNN blocks and the another one is adopted to mapping drug and gene features into one unified latent space. We believe that the fusion of those features can be more conducive to distinguishing the interaction of drug-gene pairs.

Results: Our current accuracy result is about 92%, which is higher about 5% than our previous results which published at 2018. Moreover, comparing to other two deep learning-based model, our model also achieves better accuracy, specificity and sensitivity results. As for the second experiments, ablation experiments, we try to verify the effectiveness of information fusion layers. We employed 10-fold leave-one-out cross validation method to estimate classification accuracy. The results show the classification performance of our model including information fusion layers is better than those models which only include FNN and CNN respectively.

Conclusions: We used a deep learning-based model to classify DTI prediction and two experiment results demonstrated our model can be applied as a potential classifier on drug-target interaction tasks.

## TEMPORALLY ADAPTIVE-DYNAMIC SPARSE NETWORK FOR MODELING DISEASE PROGRESSION. Zhang J, Wang Y. Arizona State University; Arizona Alzheimer's Consortium.

**Background:** Alzheimer's disease (AD) [1] is a neurodegenerative disorder with progressive impairment of memory and cognitive functions. Sparse coding (SC) [2] has been demonstrated to be an efficient and effective method for AD diagnosis and prognosis. However, previous SC methods usually focus on the baseline data while ignoring the consistent longitudinal features with strong sparsity pattern along the disease progression. Additionally, SC methods extract sparse features from image patches separately rather than learn with the dictionary atoms across the entire subject.

**Methods:** To address these two concerns, we propose a novel supervised SC network termed Temporally Adaptive-Dynamic Sparse Network (TADsNet) to uncover the sequential correlation and native subject-level codes from the longitudinal brain images. First, we adopt adaptive weights to regularize the sparse codes along longitudinal patterns of the features. Meanwhile, the adaptive structure makes it very powerful in modeling temporal sparsity patterns, especially for longitudinal data, and particularly useful in high-dimensional problems. Our approach can adaptively set various sparsities of sparse codes to minimize the errors. Secondly, we suggest that dictionary atoms should be learned on the entire subject to provide global high-level features. Thus, our approach can dynamically mine the dictionary atoms to learn the subject-level features better than patch-level features. Thirdly, taking advantage of the recurrent neural network (RNN) [3], we model the disease progression via feeding the longitudinal data into a time sequence network. Different from previous methods, our approach is a supervised time-series sparse coding, which can fully leverage the temporal and clinical patterns derived from patients' past visits. To the best of our knowledge, this is the first supervised network-based SC to model the AD progression. It adaptively adjusts the sparse codes and dynamically explores dictionary atoms on the entire subject-level in the RNN temporal learning mode.

**Results:** We study the performance of TADsNet on ADNI-1 [1] cohort (N = 3393) structural MR images and responses are the MMSE and ADAS-Cog scores, coming from baseline, 12-, 18-, and 24-months visits. The sample sizes corresponding to five time points are 837, 733, 728, 641 and 454. We use imaging data from the baseline to 24-months to predict 36-months clinical scores. We use hippocampal surface multivariate morphometry statistics (MMS) [4] as learning features. In order to evaluate the model, we randomly split the data into training and testing sets using a 9:1 ratio and repeat split 50 times to avoid data bias. We compare the proposed model with four other methods. TADsNet is our proposed supervised pipeline; TADsNet-L is unsupervised TADsNet followed by LASSO [6]; LISTA-L is LISTA [7] embedded in a simple RNN, followed by LASSO; MTSC-L [5] is the multi-task sparse coding followed by LASSO; ISTA-L is the single-task sparse coding [8] followed by LASSO. In the supervised TADsNet setting, we utilize the MSE results to guide learning sparse codes while RNN is used as a feature extractor for unsupervised TADsNet. We also compare our method with LISTA, which is a simple RNN without the adaptive-dynamic regularization. We report the comparison results for predicting 36-month MMSE and ADAS-cog scores. TADsNet and TADsNet-L outperforms all the baselines (ISTA-L and MTSC-L) by a large margin on both MMSE and ADAS-cog results and this verifies the advantages of TADsNet. Furthermore, TADsNet-L has better reconstruction power than a simple RNN based optimization method (LISTA-L) due to the adaptive and dynamic learning power. We can also notice that supervised setting of TADsNet can help improve the results of unsupervised pipeline (TADsNet-L). It may provide us the insights that the proposed algorithm has a great potential for AD diagnosis and prognosis.

**Conclusions:** In this paper, we introduce a novel supervised temporal RNN based SC model TADsNet for modeling AD progression. In our ongoing work, we integrate LSTM [9] with our model to further improve the convergence rate of TADsNet.

**DEEP MULTIMODAL BRAIN NETWORK LEARNING FOR JOINT ANALYSIS OF STRUCTURAL MORPHOMETRY AND FUNCTIONAL CONNECTIVITY.** Zhang W, Wang Y.  
Arizona State University; Arizona Alzheimer's Consortium.

Background: Current development of brain imaging acquisition, e.g., magnetic resonance imaging (MRI), allow us to noninvasively collect multimodal imaging data for the same individual with a reasonable scanning efficiency. Flourishing collections of a large amount of neuroimaging data bring challenges to a reliable data mining framework that aims to unify disparate findings from the single modality. It is widely accepted that structures and functions of the human brain have different trends and tendencies during the neurodegenerative disease progression such as autism spectrum disorder (ASD) and Alzheimer's disease (AD). Such evidences provide us an impetus to develop effective multimodal fusion methods that may benefit early intervention and biological understanding of brain disorders.

Methods: Leveraging the strong statistical power of deep models in feature learning, we design a deep graph model to achieve our fusion tasks. Specifically, we design a geometry-aware surface kernel (GSK) to extract vertex features based on the local polar coordinates. Meanwhile, we build a topology-aware network kernel (TNK) to handle brain functional connectivities and output brain regional features. These two types of features are linked through a regional pooling operation. Eventually, we can derive the graph level features for the learning tasks. Here we conduct experiments of disease classification on a relatively large brain imaging cohort, Autism Brain Imaging Data Exchange (ABIDE), to demonstrate the effectiveness of the proposed model.

Results: We report the performance of binary disease classification with accuracy and F1-scores and conduct the two-sample t-test to measure significance. Our model outperforms the baseline methods significantly. More specifically, by considering the multimodal imaging features, DMBNL achieves higher accuracy than all the single modality models, i.e. 0.25 more accurate than StrucNet and 0.16 than FuncNet. Compared to the multimodal framework (ChevbNet), our model significantly upgrades the accuracy by 0.04. Also, we observe that GSK and TNK layers are both contribute to feature learning. However, it appears the functional connectivities have more weight in the ASD prediction, i.e. missing TNK causes the larger accuracy to drop in our experiment. It is worth noting that, for all the methods, performance can be affected by the choice of the brain atlas.

Conclusions: We propose an end-to-end supervised multimodal fusion model based on the deep graph kernels. We design a unified model to automatically explore the local geometry and network connections for the multimodal brain imaging data. We validate our work in ABIDE dataset and the experimental result proves the effectiveness of our proposed model in disease prediction. To the best of our knowledge, this is the first paper that attempts to combine brain surface morphometry and functional networks via the intrinsic data properties. The proposed framework is also general and can be applied to various other neurological and psychiatric disorders.