The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Joseph Arboleda-Velasquez, M.D., Ph.D.
Assistant Professor
Schepens Eye Research Institute

"Fostering Resistance to Alzheimer's Disease Using Antibodies that Mimic the Effect of the Christchurch Variant in APOE"

Scientific Abstract
We previously reported on the characterization of a subject that resisted cognitive decline for over 30 years despite carrying the PSEN1 E280A mutation known to cause early-onset Alzheimer's. This subject was homozygote for the R136S mutation in APOE3 (Christchurch) and had lower than expected tau pathology in the presence of abundant amyloid pathology. ApoE3 Christchurch protein failed to bind to glycosaminoglycans (GAGs), a carbohydrate known to play critical roles in multiple steps of Alzheimer's pathology including amyloid formation and tau spreading. In a proof of concept experiment, a mouse monoclonal antibody raised against an APOE epitope centered around position R136 effectively blocked ApoE binding to GAGs in vitro and APOE-mediated tau pathology in mouse retinas. We hypothesize that inhibition of APOE-GAG interactions may be an effective therapy to blunt neurodegeneration in Alzheimer's disease. We propose to humanize our lead mouse monoclonal antibody as a first step towards the development of a therapeutic leveraging our discovery of the role of APOE3 Christchurch in the resistance to Alzheimer's disease. We propose the following research aims: Aim 1: To generate a panel of ApoE-GAG inhibitor human monoclonal antibodies (humAbs). Aim 2: To rank order the candidate antibodies using in vitro assays. Aim 3: To test the preclinical efficacy of two lead humAbs in mouse models of tauopathy. Completion of the proposed research is a necessary step towards future work for IND-enabling steps in the process of therapeutic antibody development.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Michelle Arkin, Ph.D.
T. William and F. J. MacWilliam Distinguished Professor and Chair of Pharmaceutical Chemistry
University Of California San Francisco Foundation

"Pharmacokinetic and Pharmacodynamic Studies of Highly Selective Caspase-6 Inhibitors in AD Models"

Scientific Abstract

Human and animal studies have implicated the protease caspase-6 (aCasp6) in the development of Alzheimer's Disease (AD). We have developed covalent Casp6 inhibitors (SU110 and SU134) that target a noncatalytic cysteine residue in aCasp6. Compounds show low nM potency in iPSC-derived neurons and high brain exposure in pharmacokinetic (PK) studies. Our current goals are to establish PK/pharmacodynamic (PD) relationships in animal models of disease. Accordingly, this 2-year project will accomplish the following aims: Aim 1. Establish biomarkers and activity of SU110 and SU134 in iPSC-derived models of familial AD. Neurons bearing TauV337M mutation express aCasp6 and caspase-cleaved Tau; inhibition of aCasp6 by SU134 reverses cell death and loss of neuronal processes. We hypothesize that mutations associated with AD, including TauP301S and APPV717I, will similarly show time-dependent expression of aCasp6 and cleaved Tau, and reversal of cell damage by treatment with SU110 and SU134. These data will inform in vivo model selection. Aim 2. Measure PK and brain exposure of Casp6 inhibitors in selected mouse model(s). We will evaluate serum and brain concentrations of SU110 and SU134 dosed PO in 5xFAD and/or PS19 mice at 4-, 7-, and 10-months of age, during disease progression. Brain tissue will be used to measure the time course for PD marker expression and for target-engagement assays. These studies will determine compound doses for subsequent PD studies. Aim 3. Establish target-engagement assays. The half-life of covalent inhibition of aCasp6 in brain will depend on target exposure and aCasp6 half-life. We will use an activity-based probe to determine the kinetics of target inhibition by SU110 and SU134 in cells and ex vivo. This assay will be used to predict dosing schedules for PD studies. Future studies will develop PK/PD relationships in 5xFAD and additional AD models. Selection of clinical candidates will follow within five years.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Harvey Cantor, M.D.
Baruj Benacerraf Professor of Immunology
Dana-Farber Cancer Institute

"Development of Engineered Brain-Penetrating Monoclonal Antibody (mAb) Targeting Osteopontin (OPN) for Alzheimer's Disease Therapy"

Scientific Abstract

Recent clinical trials of anti-amyloid-β (Aβ) antibodies have reported significant reductions in Aβ plaque load without appreciable cognitive improvement. These disappointing results may reflect a therapeutic approach that assumes that all plaques possess the same pathogenic properties. Our recent studies indicate that Aβ plaques may consist of two major subsets that include diffuse and invasive neurotoxic plaques that impair cognition, and dense plaques that represent the inert and non-toxic products of microglial processing and compaction. Both are indiscriminately targeted by current therapies.

Marked elevation of the Spp1 gene, encoding Osteopontin (OPN), by microglia is a hallmark of both animal models of AD and the human disease. Genetic deletion of OPN in 5XFAD mice substantially reduces inflammatory microglia and diffuse Aβ plaques and improves cognitive function. We have extended these findings to the human disease using clinically and neuropathologically characterized brain tissue from AD patients and controls (Mt. Sinai Brain Bank): increasing numbers of OPN-producing microglia from AD patients directly correlate with progressive dementia, according to Clinical Dementia Rating (CDR) scores.

These findings have indicated that microglial OPN drives deposition of toxic diffuse plaques, leading to neuritic destruction and cognitive impairment. This reflects a two-pronged OPN-dependent mechanism: through a) inhibition of amyloid processing and b) induction of a microglial pro-inflammatory phenotype. These considerations suggest that, in contrast to antibody-based approaches that indiscriminately target all Aβ plaques, antibody targeting of OPN may selectively reduce pathogenic plaques. We propose: 1) to define a pathogenic OPN-dependent mechanism that drives cognitive decline in the 5XFAD murine model and human AD brains and 2) to define the potential therapeutic impact of an engineered brain-penetrating antibody that targets OPN in the 5XFAD mouse model.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Se Hoon Choi, Ph.D.
Assistant Professor of Neurology
Massachusetts General Hospital

"Developing Resilient Drugs Targeting Neurogenesis and BDNF for Alzheimer's Disease"

Scientific Abstract

Alzheimer's disease (AD) destroys brain cells and synapses irreversibly, leading to dementia. A cardinal pathological lesion of the disease is the deposition of β-amyloid (Aβ). There is an urgent need for alternative strategies, as thus far, the most popular therapeutic approach aimed at reducing Aβ burden has not been proven effective in halting the disease progression. Adult hippocampal neurogenesis (AHN), a relatively novel form of brain plasticity that refers to the birth of new neurons in the adult hippocampus, remains strong in healthy human brain but drops steeply in AD patients. Yet, adult-born neurons are significantly more abundant in non-demented individuals with AD neuropathology. We previously found that inducing AHN along with elevating brain-derived neurotrophic factor (BDNF) levels provides cognitive benefits in the presence of sustained Aβ burden (Choi et al., 2018 Science). Therefore, pharmacological or natural compounds that increase AHN and BDNF (pro-AHN-BDNF compounds) might have a benefit in AD. The objective of this grant is to test the therapeutic potential of pro-AHN-BDNF compounds that we will screen and identify and that have already been reported from various animal models, using in vitro neurospheres and primary hippocampal cell cultures and in vivo AD mouse models. A successful therapy would ideally both remove the pathological hallmarks (i.e., Aβ) of AD and provide a level of functional recovery. Therefore, we will test whether co-treatment of the select pro-AHN-BDNF compounds with either γ-secretase modulator (GSM, an Aβ-reducing agent) or Aβ-antibody treatment can have a synergistic effect greater than either treatment alone. Successful completion of this proposal will generate a new therapeutic target for AD using endogenous stem cells. Leveraging the therapeutic potential of stimulating AHN and BDNF could be a new frontier and alternative for preventing or slowing down cognitive decline in AD.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Carlo Condello, Ph.D.
Assistant Professor of Neurology
University of California San Francisco

"Precision Dosing of CSF1R Inhibitors to Selectively Temper Tauopathy-Activated Microglia as a Novel Alzheimer's Disease Therapy"

Scientific Abstract

Microglia are central to Alzheimer's disease (AD) pathogenesis and have multifaceted roles in neurodegenerative processes. Drugs targeting colony-stimulating factor-1 receptor (CSF1R) to block microglial proliferation in preclinical models have shown mixed outcomes, thus the therapeutic potential of this approach remains unclear. Previsouly, we evaluated CSF1R inhibitors in tauopathy mice using multiple dosing schemes, drug analogs, and longitudinal measurements in the brain and plasma. In several models, we observed a significant reduction in tauopathy with an average of only 50-60% microglia depletion in any given experiment. Surprisingly, despite greater drug exposure and microglial depletion in male mice, we observed functional rescue and extended survival in female mice only. Notably, we observed drug dose-dependent upregulation of immediate early genes in male mice only, indicating excitotoxicity, which may have masked functional benefits. These data argue that complete microglial ablation is not required for neuroprotection and that therapeutics targeting microglia must consider sex-dependent effects on functional outcomes.

Our goal is to refine drug dosing regimens that rescue functional deficits in both sexes, whilst leaving as many healthy microglia cells intact. We will also resolve the heterogeneity of drug-dependent cellular responses to bolster our mechanistic understanding of drug action in the CNS. To increase the biological relevance and potential success of this therapeutic strategy for AD, we will also perform efficacy studies in mice that produce Aβ plaques that precede tau pathology. We hypothesize that CSF1R dosing can be precisely tuned to achieve therapeutic benefits for males and females with AD pathology. This proposal will be executed in three aims:

Aim 1: Optimizing CSF1R inhibitor dosing to maximize efficacy with least microglia cell depletion and functional rescue in both sexes.

Aim 2: Characterizing CSF1R inhibition of microglia-mediated Aβ-induced tau

pathology.

Aim 3: Deep molecular phenotyping of drug-resistant microglia and tau-laden neurons in CSF1R inhibitor studies.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Paul Greer, Ph.D.
Assistant Professor of Molecular Medicine
Eunice Kennedy Shriver Center, University of Massachusetts Medical School

"Identification of Inhibitors of MS4A Receptors"

Scientific Abstract

Alzheimer's disease (AD) is a progressive neurodegenerative disorder that exacts a devastating toll on affected individuals, their families, and our society. Despite considerable effort, existing therapeutic strategies targeting AD are limited in number and efficacy suggesting that novel approaches for treating AD are needed. Potential therapeutic targets for AD have recently come to light thanks to human genetic studies, which have revealed a relatively small number of genes whose mutation is linked to altered susceptibility to AD. Among the most compelling of these newly identified AD-associated genes are members of the Ms4a gene family, whose polymorphisms have repeatedly been shown through genome wide association studies (GWAS) to be strongly and reproducibly linked with AD. In fact, current genetic data suggest that up to 10% of all AD cases may be associated with Ms4a polymorphisms. We have recently generated exciting data showing that deletion of Ms4a genes is sufficient to rescue all behavioral and cellular phenotypes that we have examined in two different mouse models of AD. These results suggest that inhibiting Ms4a gene function is an attractive new avenue to pursue in the development of new candidate AD therapeutic strategies. Here, we propose to use two approaches to identify means of inhibiting Ms4a genes. In the first part of our proposal, we will identify small molecule chemical inhibitors of MS4A proteins using a novel, in vitro assay that we have developed. In parallel, we will take advantage of our expertise using antisense oligonucleotides (ASO) to develop ASOs that effectively inhibit Ms4a genes. Together, the two approaches described here will identify new inhibitors of Ms4a genes that can be advanced as potential therapeutic strategies for treating AD.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Daniel Lee, Ph.D.
Associate Professor
University of Kentucky

"Nutrient Sensor Modulators as Therapeutics for Alzheimer's Disease"

Scientific Abstract
To date only one disease modifying therapy for Alzheimer's disease (AD) has been approved targeting beta amyloid however treatment modalities for other phenotypes and hallmarks such as tau remain unmet in the clinic. Dysregulation of brain metabolism and slowed protein clearance increases with age and chronic conditions. Amino acid signaling impacts proteostasis but remains largely ignored as an intervention. Nutrient-sensing dysfunction offers a novel entry point for targeted therapies associated with the mechanistic target of rapamycin complex 1 (mTORC1). Arginine metabolism/ signaling becomes disrupted in AD and impacts multiple biological processes that affect both tau and amyloid deposition. Recent work uncovered lysosomal and cytoplasmic arginine sensors that regulate mTORC1 activity. GPRC6a is a G-protein coupled receptor (GPCR) that binds Lα-amino acids including arginine and may serve as an extracellular arginine sensor. Our central hypothesis argues that tauopathies promotes uncoupling of arginine-sensing in AD. Increased extracellular arginine signals "amino acid abundance" through GPRC6a to promote hyper-mTORC1 activation, which slows autophagy flux, tau clearance, and increases amyloid deposition. Genetic repression or antagonism of GPRC6a reduces receptor efficacy and signals "amino acid deficiency" to increase autophagy, tau clearance, and reduce amyloid pathology. We posit that our selective allosteric GPRC6a antagonist PF020 decreases arginine signaling to improve lysosomal function, reduces aberrant mTORC1 activity, promotes tau and amyloid clearance through autophagy. In aim 1, we will determine if PF020 decreases tau neuropathology and hallmarks of the tau phenotype in tauopathy mice. In aim 2, we will determine if PF020 decreases amyloid pathology and impacts microglial/ inflammatory signatures. We will test how PF020 impacts behavioral performance, transcriptomic signatures associated with senescence, neurotransmitter function, proteotoxic stress, and neurodegeneration. This application provides the framework for AD as a "nutrient-sensing disorder" and leverages "the first new highly selective class" of GPRC6a agents as a novel therapeutic target for AD.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Chien-liang Lin, Ph.D.
Associate Professor
The Ohio State University

"Restoration of Synapses as a Therapeutic Strategy for Alzheimer's Disease"

Scientific Abstract
Studies indicate that loss of tripartite glutamatergic synapses is the major correlate of cognitive impairment in Alzheimer's disease (AD) and occurs early in the disease process before the onset of clinical symptoms. This project is focused on restoration of synapses as a potential therapeutic strategy for AD. We have discovered and developed a novel small molecule, pyridazine-based series that can enhance the structure and function of tripartite glutamatergic synapses. We have demonstrated that our small molecules can effectively restore tripartite glutamatergic synapses and significantly improve cognitive functions in two mouse models of AD. We have developed a drug-like molecule, LH001, for clinical trial studies. We are currently conducting IND (Investigational New Drug)-enabling studies to support our IND application to the FDA for approval of human clinical trials. Once the IND application is approved by the FDA, we will begin first-in-human clinical studies in healthy subjects. However, we are currently unable to assess target engagement and monitor pharmacodynamic response in the clinical population as there is no clinically validated biomarker for LH001. It is well established that utilizing biomarker data in clinical trials significantly improves the chances of successfully completing clinical trials. The focus of this research is to identify and develop a biomarker that can assess target engagement and monitor pharmacodynamic response of LH001. Our biomarker identification approach is to identify synaptic/astrocytic proteins in plasma or CSF exosomes that are upregulated in response to compound treatment and downregulated in AD. In Year 1 (Aim 1), we will focus on identifying the LH001 biomarker candidates, validating the identified candidates, and then selecting the best candidate as LH001 biomarker. In Year 2 (Aim 2), we will focus on developing and standardizing the detection procedure.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Steven Wagner, Ph.D.
Professor in Residence
University of California San Diego

"Combination Treatment with mAducanumab and GSM-776890 in AD Mice"

Scientific Abstract

This project will determine, using two well-studied mouse models of Alzheimer's disease (AD), whether two mechanistically-distinct, amyloid-targeted and clinically-relevant therapeutic molecules, are able to be effectively utilized as a combination therapy, in order to either; a) attain greater levels of efficacy (by additive or synergistic effects), with respect to reducing cognitive impairment, attenuating neuropathological burden and lowering surrogate biomarker levels, than that which is achievable by either as a monotherapy or; b) lower the necessary efficacious doses of each when used in combination. The two therapeutic molecules are: 1) Aducanumab, an amyloid plaque-clearing immunotherapeutic human monoclonal antibody (mAb), that recently received an accelerated approval by the FDA as a treatment for AD and 2) GSM-776890, a gamma-secretase modulator (GSM), which recently demonstrated robust time- and dose-dependent efficacy in acute, sub-chronic, and chronic studies across multiple species, including primary and secondary prevention studies in a transgenic mouse model. The GSM displayed a >40-fold safety margin in rats based on a comparison of the systemic exposure (AUC) at the no observed adverse effect level (NOAEL) to the 50% effective AUC or AUC,effective, the systemic exposure required for reducing levels of $A\beta42$ in rat brain by 50%. In addition, extrapolation of rodent efficacy and toxicology data project an unprecedented 130-fold safety margin in humans based on allometric scaling and predicted human pharmacokinetics. The NIH has funded phase Ia and phase Ib clinical trials with GSM-776890, expected to begin in 2022. A recent phase III clinical trial with Aducanumab showed that in patients receiving the most effective dose (10 mg/kg), greater than 40% of those patients had complications of amyloid-related imaging anomalies (ARIA). Therefore, the possibility of the GSM, either lowering the efficacious dose of Aducanumab or replacing necessary subsequent infusions of Aducanumab, once plaques have been initially cleared, are of significant clinical relevance.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

Michael Welsh, M.D.
Professor, Internal Medicine
University of Iowa

"Developing Novel Agents that Enhance Energy Metabolism for Alzheimer's Disease"

Scientific Abstract
Alzheimer's disease (AD) is an enormous personal and public health challenge that lacks therapies that prevent progressive neurodegeneration. Identification of decreased glycolysis as a key pathogenic mechanism beginning years before symptom onset suggested that enhancing energy metabolism would be therapeutic.

We discovered that terazosin binds and activates phosphoglycerate kinase 1 (PGK1), the first ATP-generating enzyme in glycolysis. Terazosin increases ATP levels in cultured cells, mouse brain, and in preliminary studies, human brain. Stimulating PGK1 with terazosin also attenuates neurodegeneration in spinal muscular atrophy and Parkinson's disease. Preliminary epidemiologic data suggest that use of terazosin may slow AD progression in humans and may reduce tau aggregation in an AD mouse model.

Although these findings suggest that glycolytic dysfunction may be a common pathway for neurodegeneration and that enhancing PGK1 activity may have therapeutic benefit, terazosin has liabilities as an AD treatment. Terazosin was developed to inhibit alpha-1 adrenergic receptors for hypertension, and it causes orthostatic hypotension. Importantly, biochemical, functional, and structural data indicate that terazosin stimulates PGK1 independently of alpha-1 adrenergic receptor antagonism.

Our overarching goal is to enhance brain metabolic function and thereby slow AD neurodegeneration by developing new drugs that increase PGK1 activity without inhibiting the alpha-1 adrenergic receptor. Aim 1. Develop assays for agents that enhance PGK1 activity. We have three primary approaches and several secondary strategies that will allow us to identify novel chemicals stimulating PGK1. Aim 2. Develop new chemical entities to increase PGK1 activity. We will pursue complementary classical medicinal chemistry approaches and structure-based drug design. Aim 3. Test new agents that stimulate PGK1 in vivo. Depending on progress in Aims 1 and 2, we will plan in

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2021 Award Recipient

vivo tests in rodents for evaluations of safety and efficacy.

We believe this exciting strategy offers a tremendous opportunity to improve the lives of people with AD.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2019 Award Recipient

Justin Fallon, Ph.D.
Professor of Neuroscience
Brown University

Promoting Adult Hippocampal Neurogenesis as an Alzheimer's Disease Therapy

Scientific Abstract

Alzheimer's Disease (AD) is a complex disorder that is devastating for patients, their families and, if unchecked, for the medical economies in the developed world. With the recent failure of aducanumab and many other amyloid-directed therapies, it is clear that novel therapeutic targets must be explored. The hippocampus is one of the earliest and most affected brain regions in AD and its atrophy is a hallmark of disease progression. Importantly, it is now firmly established that adult hippocampal neurogenesis (AHN), which plays a major role in learning, memory and mood disorders, occurs throughout life in humans and is reduced in AD patients. Thus, promoting AHN could preserve hippocampal function and counter AD. The accumulation of negative signals that degrade the neurogenic niche are thought to be important contributors to the reduction in newborn neurons in aging and AD. BMPs negatively regulate neurogenesis and are upregulated in AD and APP transgenic mice. We recently reported that full length MuSK is a BMP co-receptor that augments and shapes BMP signaling. We have developed a mouse model that selectively perturbs MuSK-BMP signaling and thus reduces BMP 'drive'. Remarkably, these mice show enhanced AHN and improved cognition. In the proposed we will also initiate development of a candidate therapeutic that could lead to a readily translatable agent to promote AHN and improve cognition in AD patients.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2019 Award Recipient

Silvia Fossati, Ph.D.
Associate Professor of Pharmacology
Associate Director Alzheimer's Center at Temple
Temple University School of Medicine

Mitochondrial Carbonic Anhydrase Inhibitors for Alzheimer's Disease Therapy

Scientific Abstract
Mitochondria represent the energy source for brain cells, and mitochondrial damage is one of the earliest events in the development of Alzheimer's disease (AD). Preserving mitochondrial function can be a key strategy to prevent the progression of AD pathology. Carbonic anhydrases (CAs) are a family of enzymes catalyzing the conversion of CO_2 to bicarbonate and protons. CA-VA and CA-VB are in the mitochondria. We were the first to test the FDA approved CA inhibitors (CAIs) methazolamide (MTZ) and acetazolamide (ATZ) in models of amyloidosis, and to show that they are effective at preventing mitochondrial dysfunction, caspase activation and cell death in vitro and in vivo. Our preliminary data in TgSwDI mice (a model of cerebrovascular amyloidosis) confirms that the compounds are safe, and effective to reduce neurovascular degeneration and memory impairment. MTZ and ATZ, as well as most of the available CAIs, although active on both mitochondrial and cytosolic CAs, lack specificity for any of the CA isoforms. We hypothesize that the targeted inhibition of mitochondrial CAs in the brain will specifically prevent mitochondrial dysfunction and cell stress/death mechanisms induced by amyloid β (Aβ) in vitro and in vivo, ameliorating neurovascular pathology in AD models and limiting side effects.
Goals of this project are:
Aim 1. Design potent, non-toxic CAIs that are highly selective for mitochondrial CAs, improve their delivery into the brain mitochondria, and assess their metabolic stability and pharmacokinetics (PK).
Aim 2. Test this new class of compounds in our established cellular models of Aβ-mediated neuronal, glial and vascular mitochondrial dysfunction and apoptosis in comparison to ATZ, MTZ, as well as to inactive CAI analogues. Aim 3. Treat a mouse model of AD (3xTg mice) with the new CAIs (given systemically) to assess their actions on brain mitochondrial dysfunction, cell death and caspase activation in vivo.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2019 Award Recipient

Jie Gao, Ph.D.
Assistant Professor of Neuroscience
The Ohio State University

Testing Antisense Oligonucleotides Targeting IDOL-APOE Receptor Pathway as a Novel Therapy for Alzheimer's Disease

Scientific Abstract

ApoE genotype is the strongest genetic risk factor for Alzheimer's disease (AD), and has been shown to independently influence several key factors that drive pathogenesis of AD, including β-amyloid (Aβ) deposition, tauopathy, and synaptic dysfunction. In the brain, ApoE functions as a ligand for members of lipoprotein receptor family, including low-density lipoprotein receptor (LDLR), very low-density lipoprotein receptor (VLDLR), and ApoE Receptor 2 (ApoER2). Brain ApoE receptors not only regulate the metabolism of ApoE, but also mediate it's signaling pathways required for maintaining synaptic plasticity and cognitive functions. Targeting brain ApoE receptors has recently emerged as a novel therapeutic strategy to combat AD. We previously identified E3 ubiquitin ligase IDOL as a negative regulator of LDLR, VLDLR, and ApoER2 in microglia and neurons. Loss of IDOL in microglia increases LDLR protein levels, which facilitates Aβ clearance and reduces amyloid plaque burden in APP/PS1 mice, a transgenic mouse model of Aβ amyloidosis. Furthermore, loss of IDOL in neurons increases synaptic ApoER2 protein level, which has been shown to suppress tau phosphorylation and reverse ApoE4-induced synaptic dysfunction. Recently, we collaborated with Ionis Pharmaceuticals and validated an antisense oligonucleotide (ASO) targeting IDOL (hereon referred to as ''IDOL ASO''). We now propose to test whether IDOL ASO treatment can simultaneously mitigate amyloidosis, tauopathy, and ApoE4-induced synaptic dysfunction, thus targeting multiple pathological features of AD with a single therapeutic agent. The outcomes from this proposal will validate IDOL ASO as a novel therapeutic agent for the treatment of AD, provide pre-clinical evidence used to support clinical development, and serve as a guideline for future clinical study design.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2019 Award Recipient

Celeste Karch, Ph.D.
Assistant Professor of Psychiatry
Washington University in St. Louis

Novel MS4A4A-Targeting Therapeutics for Alzheimer's Disease

Scientific Abstract
The TREM2 protein controls the function of immune cells in the brain (e.g. microglia), and rare variants in TREM2 are associated with increased risk for Alzheimer's disease (AD). TREM2 likely plays a neuroprotective role in the presence of amyloid pathology by facilitating microglia activation around plaques and limiting neurotoxicity. Alternatively, TREM2 ameliorates neurodegeneration and reduces microgliosis in the presence of tau pathology. Thus, the manner and timing of targeting of TREM2 may be critical in effective treatments. We were the first to identify a major regulator of TREM2, which also modifies AD risk: MS4A4A. We found that a common allele near the MS4A4A gene (found in 30% of the population) is associated with higher TREM2 levels in the cerebrospinal fluid and is also associated with AD resilience. This suggests that targeting MS4A4A and, in turn, TREM2 could be a viable therapeutic approach for AD. These findings further suggest that TREM2 plays a role in sporadic AD and opens a new line of research that promises to revolutionize our understanding of TREM2 biology and the role of TREM2 in neurodegenerative diseases. The goal of this project is to develop antibodies that activate or inhibit MS4A4A as a mechanism to modify TREM2 function and delay AD pathogenesis. We hypothesize that MS4A4A activators will produce beneficial effects in the setting of Aβ pathology, while MS4A4A inhibitors will be neuroprotective in the setting of tau pathology. We will test this hypothesis by developing novel antibodies to activate or inhibit MS4A4A. We will then test whether MS4A4A agonists or antagonists exhibit a therapeutic benefit in the presence of amyloid or tau pathology. This proposal will yield novel biologics targeting MS4A4A and TREM2 and a better understanding of the amenability of inflammation for therapeutic targeting in AD.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2019 Award Recipient

Dianne Perez, Ph.D.
Professor
Cleveland Clinic Lerner Research Institute

Lead Optimization and Dose-Efficacy Studies of a Positive Allosteric Modulator of the Alpha1A-Adrenergic Receptor to Treat Alzheimer's Disease

Scientific Abstract

Alpha1-adrenergic receptors (ARs) regulate neurotransmission through subtypes (1A,1B,1D) known to play roles in cognitive and synaptic functions. In light of disappointing Alzheimer's Disease (AD) clinical trials focused on beta-amyloid, a novel approach is to target synaptic function.We discovered that the alpha1A-AR subtype is an AD novel target for by regulating neurogenesis and synaptic plasticity, resulting in cognitive-enhancement. We have developed a novel small molecular weight compound (comp3) that is bioavailable and is a selective positive allosteric molecule (PAM) of the alpha1A-AR. Comp3 is conformationally-selective and only modulates the norepinephrine (NE)-bound receptor but does not modulate the epinephrine (EPI)-bound receptor. Comp3 is not an agonist and does not invoke signaling on its own. Comp3 is also signaling-biased and potentiates the NE-mediated cAMP response which provides the cognitive-benefits of NE in the brain. There are no effects on the NE-mediated inositol phosphate (IP) response which causes increased blood pressure (BP) through vasoconstriction, suggesting that comp3 would not invoke a BP response. These properties of conformational and signaling-selectivity are unprecedented and would result in a drug with very little side effects. We also performed a pilot in vivo study of comp3 in an AD mouse model that showed statistically increased synaptic plasticity and cognition with no increase in BP.

This is the first report of a PAM for the alpha1-AR. It's properties would render this drug a brain-targeted modulator since NE is dominant in the brain and there is no effect of comp3 on EPI which circulates in the periphery. This drug would increase the activation of the alpha1A-AR in the brain but only when NE is being naturally released, enhancing cognition without a BP effect. Our aims are to synthesize and analyze analogs of comp3 to improve brain penetrance then to perform dose-efficacy studies using 14 mo normal rats.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2019 Award Recipient

Stephen Strittmatter, M.D., Ph.D.
Vincent Coates Professor of Neurology and Neuroscience
Yale School of Medicine

Synapse Rescue for Alzheimer's Disease by Prion Protein Antagonists

Scientific Abstract

Disease-modifying therapy for Alzheimer's disease (AD) is a massive and urgent unmet medical need. Genetic and biomarker studies demonstrate that Amyloid-ß (Aß) peptide accumulates early in AD and triggers a decades-long pathophysiology including Tau misfolding and inflammatory reaction. This process causes synapse loss and associated cognitive deficits, but the link between protein aggregation and synaptic damage had been mysterious. Moreover, therapeutic targeting of the aggregated proteins themselves has proven challenging and been unsuccessful to date. Over the last decade, we have uncovered a molecular pathway underlying synaptic damage in AD. This damage involves recruitment of inflammatory mediators, and accumulation of Tau pathology and is triggered by Aß oligomers (Aßo). More specifically, we identified the roles of cellular Prion Protein (PrPC) and metabotropic glutamate receptor 5 (mGluR5) via innovative genome-wide and synapse-wide functional screens. The unbiased nature of these discovery methods provides the highest confidence in their biological significance. It is essential to note that this approach to treating AD does not require any lowering of $A\beta$, being focused instead on the targets that drive cognitive impairment and neuronal damage.

Based on this synapse damaging pathway, we identified the synthetic polymeric compound, PSCMA, as a PrPC antagonist that is an orally available brain-penetrant. The goal of this program is to initiate a lead optimization campaign to identify discrete polymer sizes for PSCMA and PSCMA variants, and to determine the optimal ratio and order of subunits in the polymer. The intent is to maximize oral bioavailability and brain penetration while also simplifying manufacturing processes associated with polymer production on large scales. We believe this program offers an exciting opportunity to quickly and efficiently deliver a potential new therapeutic option to the clinic for a patient population in severe need of novel approaches.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2017 Award Recipient

Sandeep Datta, M.D., Ph.D.
Assistant Professor of Neurobiology
Harvard Medical School

Identifying antagonists for a novel AD risk gene

Alzheimer's disease (AD) is a devastating disorder that robs patients of their cognitive faculties, and whose prevalence is inexorably rising as our population ages. Unfortunately, currently available therapeutic agents are significantly limited both in number and effectiveness. While most efforts to develop therapeutics have remained focused on well-studied molecules such as A and Tau, geneticists have identified a host of additional genes that might be important modulators of AD pathogenesis. The Ms4a locus is amongst the best validated and most statistically significant of these hits; some estimates suggest polymorphisms in the locus may account for as many as 6 percent of late onset AD cases. We have identified the first clear molecular function for the MS4A proteins: they function as small molecule detectors. Based upon our preliminary data — which demonstrate that these proteins are highly expressed in microglia, function as receptors in microglia, are required for normal patterns of gene expression in response to inflammatory agents and Abeta peptides, are required for normal expression of microglial subtypes, and are required for normal responses to Abeta — here we propose to perform primary and secondary screens for small molecules and antibodies that act as MS4A inhibitors. These inhibitors will serve as lead compounds for potential therapeutics aimed at modulating MS4A activity in the context of neurodegenerative disease.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2017 Award Recipient

David Holtzman, M.D.
Andrew B. and Gretchen P. Jones Professor and Chairman of the Department of Neurology
Washington University in St. Louis

Development and testing of novel TREM2-targeted therapeutics

The high-profile failure of numerous amyloid-targeting therapies in trials of symptomatic AD patients indicates the need to treat individuals in the early, pre-clinical phase of the disease as well as the importance of developing treatments targeting disease processes downstream of amyloid pathology, such as inflammation. Rare coding variants in TREM2 increase the risk of developing AD by 2-4 fold, although the functional consequences of the variants and the role of TREM2 in AD are unclear. Several studies indicate that TREM2 is critical for microglial responses to inflammatory brain insults, including $A\beta$ plaque deposition. APPPS1 mice expressing only one or no copies of Trem2 exhibit reduced plaque-associated microgliosis and a concomitant increase in plaque-associated dystrophic neurites. These results suggest that TREM2 serves a neuroprotective role in the presence of amyloid pathology by facilitating microglial activation around plaques to limit plaque-associated neurotoxicity. However, our unpublished data examining the role of TREM2 in tau pathology found that Trem2-deficiency ameliorates neurodegeneration, and reduces microgliosis in the P301S mouse model of tauopathy. Thus, whether TREM2-dependent microglial functions could be beneficial or harmful might depend on the particular stage of AD. The goal of this proposal is to develop antibodies that either activate or inhibit TREM2 (Aim 1) and test whether TREM2 agonists or antagonists exhibit a therapeutic benefit in the presence of amyloid or tau pathology as a preclinical, proof-of-principle study (Aim 2). We hypothesize that TREM2 agonists will elicit beneficial effects on neuritic dystrophy in the setting of $A\beta$ pathology, while TREM2 antagonists will be neuroprotective in the setting of tauopathy. The outcomes from this proposal will yield novel biologics targeting TREM2 and a better understanding of the amenability of TREM2 and TREM2-dependent inflammation for therapeutic targeting in AD.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2017 Award Recipient

Kenneth Kosik, M.D.
Harriman Professor of Neuroscience Research
University of California Santa Barbara

Farnesyltransferase inhibitors to Treat Neurofibrillary Pathology of Alzheimer's Disease

The aims proposed here will lay the groundwork for advancing the very promising preliminary data toward a clinical trail to treat the primary tauopathies. Farnesyl transferase inhibition using the drug lonafarnib via a target identified as the farnesylated protein Rhes, a member of the Ras-GTPase family has striking effects on tau pathology.
Inhibition of Rhes can prevent behavioral changes, brain shrinkage, formation of tau inclusions, microgliosis, and widespread sumoylation and ubiquitination in mice that harbor a tau mutation. This proposal will focus on systems pharmacology and mechanistic questions that will track in detail the pathway from farnesyl transferase inhibition to Rhes suppression to the amelioration of tau pathology We hypothesize that Rhes pathway detection and removal of aberrant tau moieties operates through autophagy and sumoylation. Extensive human experience with lonafarnib for unrelated medical conditions will greatly ease rapid repurposing of this drug for patients. The long term objectives are to show that Rhes pathway regulation through farnesylation can prevent neurofibrillary disease and lonafarnib can be re-purposed to treat tauopathies.

The aims pose the following questions: Does treatment with Lonafarnib alters the levels of farnesyl-Rhes and or the farnesylation of lysosomal proteins. Does tau interact directly with Rhes? What are the immediate downstream components in the Rhes pathway that favor a reduction in tau pathology? What forms of autophagy are activated by Rhes in the presence of tau mutations? What is the molecular nature of the sensor that first detects mutant tau? What is the role for Sumoylation in the tauopathies and the Rhes pathway?

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2017 Award Recipient

Seulki Lee, Ph.D.
Assistant Professor of Radiology
Johns Hopkins University School of Medicine

A Novel GLP-1 Receptor Agonist for the Treatment of Alzheimer's Disease

Recently, it was suggested that activation of microglia leads to the conversion of resting astrocytes to A1 astrocytes in various neurodegenerative diseases including Alzheimer's disease (AD). In AD, the abnormal misfolding and aggregation of beta-amyloid (Ab) induces toxic effects on neurons. In our preliminary study, we found that Ab and a-synuclein aggregates induce M1 microglial activation and facilitate A1 astrocyte formation by secreting TNFa, IL-1a, IL-1b, and C1q (reported as A1 astrocyte inducer), resulting in the death of primary human neurons. Therefore, the development of agents that could inhibit the formation of M1 microglia and A1 astrocytes without off-target toxicity could have profound therapeutic potential in the treatment of neurodegenerative disorders for which there currently are not disease modifying therapies.

NLY01 is a differentiated, CNS penetrating, long-acting GLP-1R agonist with extended half-life in monkeys. We found that NLY01 selectively inhibits Ab-induced microglial activation through upregulated GLP-1Rs and blocks A1 astrocyte formation, thus protecting neurons. Importantly, in a small number of transgenic (Tg) animal models of AD, NLY01 is profoundly neuroprotective against the 3xTg-AD models and restores memory functions via targeting microglial-mediated conversion of astrocytes to neurotoxic A1 astrocytes.

We aim to explore the pharmacological efficacy of NLY01 in greater detail towards direct-to-Phase 2 clinical trials in AD patients. In Aim 1, we aim to initiate a dose-dependent efficacy study of NLY01 and determine its PK/PD profiles as well as its ability to penetrate the brain in a complementary AD model, 5xFAD Tg AD mice, to drive clinical translation. In Aim 2, we will develop a clinical trial protocol for AD patients with defined primary and secondary endpoints and submit a pre-IND package to the FDA. With detailed evidence of NLY01's therapeutic potential in AD based on studies proposed here, we will plan proof of concept clinical studies in AD patients.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2017 Award Recipient

Chien-liang Lin, Ph.D.
Associate Professor
The Ohio State University

Restoration of tripartite glutamaterigic synapse as a therapeutic strategy for Alzheimer's disease

Mounting evidence indicates that glutamate dyshomeostasis plays a crucial role in the pathogenesis of Alzheimer's disease (AD). Glutamate transporter EAAT2 plays a critical role in the homeostatic regulation of extracellular glutamate levels. EAAT2 also plays an essential role in cognitive memory functions. However, loss of EAAT2 protein and function are commonly found in AD patients and are an early event in disease pathology. We have discovered a series of novel compounds that can increase EAAT2 protein expression via a novel translational activation mechanism. We have demonstrated that our compounds can significantly improve cognitive functions and restore synaptic integrity in both APP and tau mouse models of AD. This project is currently at the clinical candidate selection phase. The goal of this study is to determine a clinical candidate and then move forward to IND-enabling studies.

The Edward N. & Della L. Thome Memorial Foundation Awards Program in Alzheimer's Disease Drug Discovery Research
2017 Award Recipient

Thomas Wisniewski, M.D.
Lulu P. and David J. Levidow Professor of Neurology;Professor of Neurology, Pathology and Psychiatry
New York University School of Medicine

Developing Peptoid Inhibitors to Target the ApoE/Aβ Interaction as a Novel Alzheimer's Disease Therapy

The pathological accumulation of Aβ peptides as toxic oligomers, amyloid plaques and cerebral amyloid angiopathy (CAA), either from increased production of Aβ peptides or from their inadequate clearance, is critical in the pathogenesis of Alzheimer's disease (AD). The apolipoprotein E4 (apoE4) allele, a major genetic risk factor for late-onset AD, has been identified and strongly associated with increased amyloid plaques deposition in brain parenchyma and advanced vascular amyloid pathology such as CAA. Numerous studies have shown that apoE binds to residues 12-28 of Aβ and this binding modulates Aβ accumulation hence affecting disease progression. However, there is no consensus on how different apoE genotypes contribute to the pathogenesis of AD. We suggest that the optimal therapeutic strategy may be to specifically prevent the interaction of apoE with Aβ, allowing us to circumvent complications of altering apoE levels which may cause detrimental effects on the many other beneficial roles apoE plays in neurobiology. Recently, using peptidomimetic technology and the macrocyclization method, we designed a peptoid library derived from Aβ12-28P sequence to screen for new apoE/Aβ binding inhibitors with higher efficacy and safety for blocking the apoE/Aβ interaction. Peptoids are N-substituted glycine oligomers, which recapitulate many desirable attributes of natural peptides including formation of stable secondary structures and demonstration of a range of biological activities. Our Specific Aims are to:

1) Design and in vitro characterization of non-toxic, pharmacokinetically favorable peptoid antagonists of the apoE/Aβ interaction.
2) The lead peptoid will be tested in vivo using 3xTg mice and TgSwDI mice crossed onto human apoE3 or E4 or knock out (KO) backgrounds.

Karen Ashe, M.D.
Chair, Neurology and Neuroscience, University of Minnesota Medical School

Discovery of Caspase-2 Inhibitors to Treat Dementia

Caspase-2 inhibitors have excellent, though unrealized, potential for improving cognition in patients with Alzheimer's (AD), frontotemporal dementia (FTD), and Huntington's disease (HD), based on the restorative effects of lowering caspase-2 in mouse models of these diseases. Our broad, long-term objective is to prepare safe and effective caspase-2 inhibitors to treat dementia. We recently discovered that caspase-2 forms a toxic tau fragment (Δtau314) in cell and mouse models of FTD. Δtau314 infiltrates dendritic spines and dislocates glutamate receptors, impairing synaptic function. Lowering caspase-2 in cells prevents tau from infiltrating spines, and reducing caspase-2 in the brain restores memory in impaired mice. We will employ this animal model to provide confidence in mechanism for our inhibitors, in order to exploit using Δtau314, the only known molecular biomarker of caspase-2 activity that is correlated with changes in cognition. Caspase-2 inhibitors have been reported but none are both selective and brain-penetrant. The failure of these compounds to advance to the clinic suggests the need for a radical shift in drug discovery paradigms. What is required is a paradigm that is built upon holistic assays (those that account for the complex interactions between caspases and intracellular components), rather than the conventional drug discovery approach based on assays employing recombinant enzymes. We propose to establish a holistic platform for the discovery of selective, safe, and effective caspase-2 inhibitors. We plan to accomplish this by pursuing the following specific aims: (1) preparation and assay of compounds inspired by Δtau314, the canonical inhibitor VDVAD-CHO (D-CHO = aspart-1-al), and other reported brain penetrant or selective caspase-2 inhibitors, (2) development of holistic assays to study the caspase activity and selectivity of compounds in brain cell lysates, and (3) mouse PK/PD (oral, sc, ip) studies on the best of these compounds to characterize their brain-distribution and ability to restore cognitive function.

Yueming Li, Ph.D.
Associate Member/Professor, Memorial Sloan-Kettering Cancer Center

Development of TFEB target-based small molecules for Alzheimer's disease therapy

The overall objective of this proposal is to develop small molecules that promote TFEB-mediated clearance of misfolded Tau proteins for treatment of Alzheimer's disease (AD). Aberrant Tau phosphorylation, leading to the formation of intracellular neurofibrillary tangles (NFT) found in AD patient brains, is a hallmark of AD. The Transcription Factor EB (TFEB) was discovered as a master regulator for lysosomal biogenesis and autophagy. Our studies have shown that mild overexpression of TFEB potently reduces tauopathy, behavioral deficit and neurodegeneration in a Tau transgenic mouse model by activating autophagy and lysosomal degradation pathways indicating that upregulation of TFEB could be a promising strategy for AD therapy. In this application, we propose to first conduct a large-scale small molecule library screening to identify hits that target the TFEB pathway and generate lead compounds. Secondly, we will determine the therapeutic potential of the lead compounds in AD preclinical models. The proposed research consisting of chemical library screening, validation and analog synthesis, and in vivo testing will enable us to develop drug candidates that are effective in clearing the Tau/NFT pathologies for AD treatment.

Susan Lindquist, Ph.D.
Member, Professor of Biology, Whitehead Institute for Biomedical Research

Bioactive Cyclic Peptides as Potential Therapeutics for Alzheimer's Disease

Alzheimer's disease (AD) is characterized by toxic conformations of Amyloid-beta (Abeta) peptides. A poorly understood mechanism of Abeta's toxic effect in neurons together with the lack of high-throughput screening systems against Abeta cytotoxicity has made the discovery of AD therapeutics difficult and painstakingly slow.

Exploiting the extraordinarily conserved pathways of protein trafficking and homeostasis in eukaryotes, we established powerful Abeta42 cytotoxic models anchored at one end by high-throughput yeast screens and validated at the other by human stem cell-derived neurons. Genome-wide screening in yeast identified genes as modulators of critical cellular pathways disrupted by Abeta toxicity. Importantly, human homologs of several of these genes are validated risk factors for AD (i.e., PICALM), thus corroborating the pathobiological relevance. Collaborating with the Harvard Institute of Chemistry and Cell Biology and the National Center for the Advancement of Translational Sciences, we screened >500,000 compounds and identified hit compounds (~150) that reverse toxicity in yeast. These compounds have now been licensed to Yumanity Therapeutics (http://yumanity.com) to ensure that their potential therapeutic applications are adequately explored. (Previous research on two of these compounds in the Lindquist laboratory was supported by the Thome Foundation.)

We have established additional cytotoxic models in yeast and neurons by expressing familial Abeta variants. Intriguingly, the familial Arctic mutation (E22G) strongly increased cytotoxicity. Remarkably, a non-toxic mutant, I31E, abrogates cytotoxicity of toxic Abeta variants in coexpression systems, emphasizing the importance of peptide biologics as potential therapeutics.

Recently, the therapeutic potential of biologics in medicine has begun to materialize. The early promise of the phase 3 clinical trial for aducanumab (Biogen Idec) as an antibody biologic is a ray of hope for AD therapy. Using our powerful cytotoxic models, we propose to isolate, characterize and validate cyclic peptides as potential AD therapeutics. These peptide biologics provide a novel paradigm for drug development.

Kun Ping Lu, M.D., Ph.D.

Professor of Medicine, Beth Israel Deaconess Medical Center

Development of Novel Targeted Therapy for Alzheimer's Disease

Prevalence of Alzheimer's disease (AD) may quadruple worldwide by 2050, but effective treatment is not available. Tauopathy made of hyperphosphorylated tau is one hallmark lesion in AD. Immunization against tauopathy epitopes shows promising efficacy in mouse models. Tauopathy correlates well with memory decline in AD and is also a defining feature of other tauopathies, notably chronic traumatic encephalopathy (CTE). Significantly, one of the best-known environmental risk factors for CTE and AD is traumatic brain injury (TBI). However, tauopathy is not obvious in acute and subacute TBI and how TBI leads to tauopathy is unknown.

We have identified a unique proline isomerase Pin1 to inhibit tauopathy by converting the phosphorylated Thr231-Pro motif in tau (p-tau) from cis to trans conformation. By generating cis and trans p-tau antibodies, we have identified the early pathogenic cis tau leading to tauopathy in AD. We have now created high affinity monoclonal antibody (mAb) that effectively removes this early, secreted and toxic cis tau in vitro and in mice. Importantly, cis p-tau is an early precursor of tauopathy and an early driver of neurodegeneration that can be blocked by cis mAb. Our data provide a direct link from TBI to CTE and AD, and suggest that cis mAb may be further developed for early diagnosis and treatment of AD, TBI and CTE.

Here we will first humanize our cis p-tau mouse mAb and then characterize humanized mAbs and evaluate their potency to eliminate cis p-tau and inhibit cistauosis in vitro as well as to treat tauopathy in AD mouse models. The current proposal is the first and essential step of our ultimate goal to develop unique therapeutic mAb against the disease-driving early pathogenic tau for treating AD, raising the unique opportunity of halting or preventing tauopathy and memory loss in AD patients at early stages.

Pamela Maher, Ph.D.
Senior Staff Scientist, The Salk Institute

Novel Glycation Inhibitors for the Treatment of Alzheimer's Disease

Age is the single greatest risk factor for Alzheimer's disease (AD). Among the pathophysiological changes that have been proposed to contribute to the decrease in brain function with age are increases in protein glycation. Protein glycation, the non-enzymatic addition of sugars to proteins, results in the formation of advanced glycation end-products (AGEs). This protein modification is increased during normal aging and is greatly exacerbated in AD suggesting that inhibitors of AGE formation may have potential for the treatment of AD. Glutathione (GSH), the major endogenous antioxidant, plays a critical role in the removal of the potent AGE precursor, methylglyoxal (MG). Loss of GSH occurs during aging suggesting that GSH maintenance should be an integral part of any approach to preventing the accumulation of AGEs. Fisetin was initially identified as an orally active, novel neuroprotective and cognition-enhancing molecule. Fisetin can protect nerve cells from multiple toxic insults and can increase the intracellular levels of GSH in the presence of oxidative stress. Furthermore, fisetin can inhibit protein glycation both in vitro and in vivo. We recently synthesized a series of much more potent fisetin derivatives, many of which maintain the ability to increase GSH levels. These derivatives have the potential to be good CNS drugs and do not suffer from the intellectual property challenges of the natural product fisetin. Based on these observations, it is proposed to advance fisetin derivatives as inhibitors of AGE accumulation for the treatment of AD. Specifically, the anti-glycation activity of the fisetin derivatives will be assayed in the test tube and in cell culture models of glycation. The most effective of these derivatives will be subjected to limited pharmacokinetic and toxicology studies. Based on these results, the two best derivatives will be tested in SAMP8 mice, a novel animal model of aging and AD.

Benjamin Wolozin, M.D., Ph.D.
Professor, Boston University School of Medicine

*Targeting RNA Metabolism and the Stress Granule Pathway to Inhibit Tau
Aggregation*

RNA binding proteins (RBPs) use physiological aggregation to form inclusions composed of protein/RNA complexes that control of processes such as RNA transport, RNA translation and RNA degradation. For instance, stress causes RBPs to exit the nucleus where they form stress granules, which function to sequester away from the pool of translated transcripts facilitating translation of protective proteins.

We have discovered that RBPs, such as TIA1, co-localize with tau pathology. Further work shows that RBPs, such as TIA1, are important regulators of tau pathology. Knockdown or knockout of TIA1 inhibits tau (WT or P301L) aggregation and toxicity in neurons in vitro and in vivo, while TIA1 over-expression stimulates tau aggregation and toxicity. In Alzheimer's disease, this pathway becomes hyperactive. The intimate link between tau, RBPs and neurodegeneration points to novel approaches to pharmacotherapy of tauopathies. RNA granules, including stress granules, are regulated by signaling networks. These networks can be pharmacologically modulated to reverse formation of both tau and RNA granules.

In this proposal our goal is to identify compounds that can reverse tau aggregation and toxicity associated with tau/TIA1 expression. We have generated a line of neuronal cells (human SY5Y cells) that inducibly express both tau and TIA1. Expressing both proteins together induces aggregated tau, tau inclusions and degeneration. In Aim 1 we will screen our tau/TIA1 cell line against a library of compounds selected for likely CNS penetration based on Lapinski's rules. Two phenotypes will be selected as outcomes: Tau granule formation and neurotoxicity. Hits will be validated with dose response curves and then in assays of primary neuronal cultures expressing P301L tau. Tertiary validation will use biochemistry. Lead compounds will be tested for future studies in vivo. In Aim 2 we will investigate the ADME and pharmacokinetic properties of the most promising hits.

P. Jeffrey Conn, Ph.D.

Lee E. Limbird Professor of Pharmacology; Director, Vanderbilt Center for Neuroscience Drug Discovery, Vanderbilt University Medical Center

In vivo characterization of metabotropic glutamate receptor subtype 5 positive allosteric modulators in a mouse model of Alzheimer's disease

Alzheimer disease (AD) is the most common form of dementia and is characterized by the progressive decline in cognitive function, with the primary deficits being hippocampal-mediated learning and memory loss. Recent studies suggest the involvement of glutamate in the pathology of the disease, as levels are decreased in the hippocampus of AD patients. Glutamate modulates excitatory postsynaptic currents via metabotropic glutamate receptors (mGlus). mGlu5 is the most highly expressed mGlu in the hippocampus and a close signaling partner of the N-Methyl-D-aspartate receptor (NMDAR). The NMDAR is critical in regulating hippocampal synaptic plasticity and essential for hippocampal-dependent cognitive function. Therefore, increased activation of mGlu5 offers an exciting new therapeutic strategy to enhance cognitive function in patients suffering from AD. Recently, our group has developed a highly potent, selective series of mGlu5 positive allosteric modulators (PAMs) with enhanced pharmacokinetic properties for in vivo studies, providing an unprecedented opportunity to evaluate the potential of selective potentiation of mGlu5 as a novel target for the treatment of symptoms associated with AD. Unlike orthosteric agonists, PAMs dramatically potentiate the receptor response to its endogenous ligand glutamate offering high selectivity while avoiding unwanted side-effects seen with direct activation. CK-p25 mice have a loss of hippocampal synaptic function as well as cognitive function, In addition, these mice exhibit the hallmark pathological and neurodegenerative features of AD and allow the opportunity to characterize the ability of our novel mGlu5 PAMs to restore cognitive deficits in a preclinical animal model that best emulates the human disease state. Studies proposed will establish the ability of mGlu5 PAMs to restore deficits in synaptic function, determine the degree of in vivo occupancy of central mGlu5 necessary to observe in vivo efficacy and evaluate the cognitive-enhancing efficacy of mGlu5 PAMs in preclinical models of cognitive function in an animal model of AD.

Philip De Jager, M.D., Ph.D.

Associate Professor
Harvard Medical School

Identification of small molecules that modify CD33 expression

With the discovery and validation of Alzheimer's disease (AD) susceptibility loci, we now have AD risk factors that give insights into the earliest pathophysiological processes of AD. Specifically, recent genome-wide studies have identified nine non-APOE AD susceptibility loci: ABCA7, BIN1, CD2AP, CD33, CLU, CR1, EPHA1, MS4A, and PICALM. Several of these loci implicate the innate immune system in susceptibility to AD. Here, we focus on the CD33 locus and propose to identify compounds that block the functional consequences of the CD33 allele that increases an individual's risk for AD. The proposal leverages the detailed functional characterization of this risk allele that we have conducted and highlights a disturbance in monocyte function that could play a broader role in AD susceptibility, beyond its mediation of the effect of a risk allele. CD33, an inhibitory molecule, expressed on the surface of myeloid progenitors, monocytes and macrophages that constitutively reduces the activity of myeloid cells. Our investigations have uncovered a robust molecular phenotype associated with the CD33 risk allele: it increases the surface expression of CD33 6-fold on circulating monocytes. This large effect on the state of activation of monocytes is present both in younger and older subjects, suggesting that the functional consequences of the CD33 locus may be exerted from the earliest stages of AD pathophysiology, which probably occur in middle age. We hypothesize that altered myeloid function driven by high CD33 expression contributes to the onset of AD, and therefore propose to identify chemical agents, using high throughput screening, that reduce CD33 surface concentration on monocytes of subjects with the risk allele to the levels seen on monocytes from subjects of the protective allele.

David Harris, M.D., Ph.D.
Professor and Chair
Boston University School of Medicine

Prion Protein-Targeted Therapeutics for Alzheimer's Disease

The purpose of this application is to develop an entirely new class of therapeutic agents for Alzheimer's disease, based on the recent identification of a novel drug target, the cellular prion protein (PrPC), which functions as a receptor for Abeta oligomers and may mediate their pathogenic effects. Small molecule ligands that bind to PrPC and block its interaction with Abeta oligomers may therefore represent powerful drugs to prevent neuronal and synaptic dysfunction in Alzheimer's disease.

We have already identified several lead compounds that bind specifically and with high affinity to PrPC, and inhibit its interaction with Abeta oligomers, thus demonstrating unequivocally that PrPC is a druggable target. To accomplish this, we used in silico methods to predict probable ligand binding sites on the surface of PrPC, followed by a virtual screen of 17 million compounds to identify those that docked optimally in one of these sites. Candidate ligands were then validated experimentally using biophysical techniques to measure their affinity for PrPC, and methods developed by my laboratory to measure their effects on the functional activity of PrPC. Our most promising set of lead compounds bind tightly and specifically to PrPC and powerfully inhibit its ability to bind to Abeta oligomers.

We propose here to: (1) characterize at the atomic level the site on PrPC to which our lead compounds bind; (2) optimize the molecular scaffold of these compounds using structure-activity analyses and structure-based design methods; and (3) test whether these compounds and their derivatives suppress the synaptotoxic effects of Abeta oligomers in cellular and animal models.

Susan Lindquist, Ph.D.

Member, Whitehead Institute
Full Professor of Biology, Whitehead Institute for Biomedical Research

A Yeast Model of Abeta Toxicity for Drug Discovery

The Abeta peptide is a central player in Alzheimer's Disease (AD). Abeta is processed from the full length Amyloid Precursor Protein and populates large plaques throughout the brain. However, smaller oligomeric species are widely believed to cause cell death. Unfortunately, efforts to reduce Abeta processing or promoting clearance have largely failed. We have thus created a much simpler model of Abeta toxicity for unbiased phenotypic screens free of prejudice about mechanism. To this end, we use the budding yeast, Saccharomyces cerevisiae, to capture agents that reduced Abeta toxicity. Though lacking the complexities of a nervous system, yeast offer nearly all of the conserved cellular pathways involved in most aspects of basic eukaryotic cell biology, including the sophisticated protein homeostasis mechanisms that cope with the cellular stresses imposed by toxic neurodegenerative disease proteins. In the yeast model of Abeta toxicity, the peptide is targeted to the endoplasmic reticulum and samples the secretory pathway. A genetic screen against Abeta toxicity identified the yeast homolog of PICALM, a risk factor for AD in humans. We validated genetic modifiers in both a C. elegans model and an Abeta oligomer assay in rat neuronal cultures. For this proposal, we have one completed and one ongoing phenotypic drug screen for compounds that combat Abeta toxicity. Importantly, we identified the AD-relevant compound clioquinol (CQ), which rescues Aβ toxicity and cognition in a mouse model of AD. A close derivative of this compound has shown promise in early clinical trials. Here, we propose to enter the compounds that reduce Abeta toxicity into a pipeline of secondary screens, neuronal assays, and medicinal chemistry. We will partner with chemist Stephen Buchwald (MIT) to perform extensive structura activity relationships. We propose to structurally and mechanistically characterize several compounds protective against AΒ toxicity, taking advantage of yeast genetics and cell biology.

David Morgan, Ph.D.

Distinguished Professor and Executive Director
University of South Florida College of Medicine

Histone deacetylase-6 inhibition as a treatment for tauopathy

Deposition of the microtubule associated protein tau is associated with a number of neurodegenerative disorders. The most common tauopathy is Alzheimer's disease, which contains both tau deposits and amyloid deposits in affected brain regions. To date, there are no effective treatments targeting tau deposition.

Histone deacetylase 6 (HDAC6) is an enzyme that removes acetyl groups from lysine containing proteins. Two substrates of HDAC6 are acetylated tubulin and acetylated heat shock protein 90 (HSP90). Histone proteins do not appear to be substrates for this family member. HSP90 is a chaperone that is a decision point in the life cycle of misfolded proteins, leading to either refolding or to degradation. Tau is one of the clients served by HSP90. Recent work by the Petrucelli group indicates that acetylation of HSP90 shifts the balance towards degradation. Thus inhibition of HDAC6 leads to an increase in the acetylated form of HSP90 and increased degradation of its client proteins, including tau.

We have tested a selective HDAC6 inhibitor tubastatin A in a mouse model of tau deposition (Tg4510). We found that daily injections of tubastatin rescue memory deficits in these mice, and reduce total tau, but not phsophorylated forms, in the mouse brain. These data suggest that selective HDAC6 inhibitors may be useful therapeutic approaches for the treatment of Alzheimer's and other tauopathies.

To further test this hypothesis, we propose 2 aims:
Aim 1. Optimizing tubastatin dose and administration. We will treat mice with multiple doses an measure brain an blood concentrations and accumulation of acetylated HDAC6 targets using oral administration.

Aim 2. Time course of tubastatin effects on the phenotype of Tg45410 mice. This aim will test whether initiating treatment early will demonstrate greater protection and if the rescue of the phenotype can be sustained over time.

Luigi Puglielli, M.D., Ph.D.
Associate Professor
University of Wisconsin-Madison

ATase1/ATase2 inhibitors for the prevention of Alzheimer's disease

Our group has identified a novel form of post-translational regulation that affects both levels and activity of BACE1. Specifically, we discovered that nascent BACE1 is transiently acetylated in the lumen of the ER by two acetyltransferases, which we named ATase1 and ATase2. The acetylated intermediates of nascent BACE1 are able to complete maturation whereas non-acetylated intermediates are rapidly degraded. Consistently, up-regulation of ATase1 and ATase2 increases BACE1 levels and Abeta generation while down-regulation has the opposite effects. Both ATase1 and ATase2 are preferentially expressed in neurons and are up-regulated in the brain of late-onset AD patients. Following up on our initial discovery, we have now identified novel biochemical inhibitors of ATase1 and ATase2 that significantly reduce the levels of BACE1 and the generation of Abeta in cellular systems. The mechanism of action of the compounds involves competitive and non-competitive inhibition as well as generation of unstable intermediates of the ATases that undergo degradation. Here, we report the completion of the physical/chemical (pre-formulation) characterization and formulation development phase and successful transition to animal models of the disease. The initial results in AD animal models show successful changes in BACE1 and APP metabolism as well as prevention of the early deficits that characterize the AD-like pathology of the animals. The general hypothesis of this research is that biochemical inhibitors of ATase1 and ATase2 can serve to prevent or delay AD dementia. Specific Aim 1 will assess whether recently identified compounds that inhibit ATase1 and ATase2 activity can delay or block AD-like neuropathology in animal models of the disease. Specific Aim 2 will characterize the biochemical properties as well as mechanism of action of two new active compounds that inhibit ATase1 and ATase2 in vitro and reduce BACE1 levels in cell-based settings.