Vascular Contributions to Dementia: Modeling the Disease and Hope for Therapies

M. Paul Murphy, Ph.D.
University of Kentucky
Molecular and Cellular Biochemistry
Sanders-Brown Center on Aging

Disclosure

I, M. Paul Murphy, DO NOT have a financial interest/arrangement or affiliation with one or more organizations that could be perceived as a real or apparent conflict of interest in the context of the subject of this presentation.

This research was supported by the following funding organizations: NIH (NINDS, NIA, NIDDK, NIGM, NIEHS), NSF, AFAR, CART, Alzheimer’s Association, American Heart Association, and the Bright Focus Foundation

Rachel Ahmed, Grad Student
Robin Webb, Grad Student
Chloe Malley, Grad Student
Thomas Platt, Grad Student
Ann Helman, Grad Student
Valerie Roven, Post Doc
Christa Studzinski, Post Doc
Katie McCarty, Research Technician
Katharine Kehoe, Research Technician
Tina Berkolt, Staff Scientist
Dana Niedowicz, Staff Scientist
Terese Macheda, Staff Scientist
Alzheimer’s Disease

- Most common neurodegenerative condition in the elderly
- In addition to neuronal loss, there are two major lesions in AD:
  - Extracellular, neuritic plaques (A)
  - Intracellular, neurofibrillary tangles (C)
- Tangles appear first, and are a better correlate of dementia

Neurodegenerative Disease

<table>
<thead>
<tr>
<th>Age</th>
<th>Incidence</th>
</tr>
</thead>
<tbody>
<tr>
<td>60</td>
<td>&lt;1%</td>
</tr>
<tr>
<td>65</td>
<td>~5%</td>
</tr>
<tr>
<td>75</td>
<td>~10%</td>
</tr>
<tr>
<td>85</td>
<td>~20%</td>
</tr>
<tr>
<td>95</td>
<td>~40%?</td>
</tr>
</tbody>
</table>

AD is the Most Common Cause of Dementia in the Elderly

- Alzheimer’s disease
- Cerebrovascular disease
- Lewy body disease
- Frontotemporal dementia

Age at Death

Institutionalization

Institutionalization

Less Serious Health Issue

Partially Effective Therapy

Partially Effective Therapy

Slowed Rate of Decline

Slowed Rate of Decline

Alzheimer’s Disease

- Most common neurodegenerative condition in the elderly
- In addition to neuronal loss, there are two major lesions in AD:
  - Extracellular, neuritic plaques (A)
  - Intracellular, neurofibrillary tangles (C)
- Tangles appear first, and are a better correlate of dementia

Age at Death

Institutionalization

Institutionalization

Less Serious Health Issue

Partially Effective Therapy

Partially Effective Therapy

Slowed Rate of Decline

Slowed Rate of Decline

Alzheimer’s Disease

- Most common neurodegenerative condition in the elderly
- In addition to neuronal loss, there are two major lesions in AD:
  - Extracellular, neuritic plaques (A)
  - Intracellular, neurofibrillary tangles (C)
- Tangles appear first, and are a better correlate of dementia
Amyloid Pathology is Generic

Adapted from: Forman et. al., Nature Medicine, 10: 1055-63, 2004

AD (Plaque) FTD (Tangle) PD (Lewy Body)

ALS (Cytoplasm Inclusion) HD (Nuclear Inclusion) CJD (PrP res Plaques)

Plaques and Tangles: Causal or Markers?

Plaques (Aβ) Tangles (tau)

Neuronal Dystrophy and Death

Dementia

Aging Diet & Metabolism Pathogenic Mutations Genetic Modifiers

APP

Toxic Aβ Oligomer Plaques

Tangles
Plaques and Aβ

- Aβ is generated from the Amyloid Precursor Protein (APP)
- About 90% of the Aβ generated is 40 amino acids long (Aβ40), and is relatively soluble in aqueous solution.
- Another ~5-10% is slightly longer (Aβ42), and not very soluble: biophysical studies have consistently shown that Aβ42 is prone to aggregation.

Amyloid Precursor Protein (APP) Processing

Amyloidogenic APP Processing
Is AD a loss of Repression on the Aβ Producing Enzymes?

- **Normal Aging**
  - β-Secretase
  - γ-Secretase

- **Pathologic Aging**
  - β-Secretase
  - γ-Secretase

**Aβ**

- Healthy Brain
- Alzheimer's Disease

Adapted from: Roberson & Mucke, Science, 314: 781-784, 2006
β-Secretase

- BACE (β-secretase) is the rate limiting enzymatic step in the production of Aβ.
- An increase in β-secretase activity occurs in AD, but the reason is unknown.
- It is possible that increasing β-secretase activity accounts for a substantial portion of sporadic AD cases.

BACE1 in Neurons, BACE2 in Neurons and Astrocytes

Cerebrovascular Disease is the Second Most Common Cause of Dementia
What is Vascular Dementia?

AD + Cerebrovascular Disease Is an Exceptionally Common Occurrence

Dementia, Obesity and Diabetes
Diet, Metabolism and Modulation Studies in Mice

Known:
- Type 2 Diabetes Mellitus confers increased risk of AD
  - Unknown mechanism(s)
- High fat Western diets increase Aβ accumulation
- Caloric restriction reduces Aβ levels
- Oxidative stress and inflammation are involved in both diseases.

Experiments:
- APP knock-In Mice
- APP Over Expressing Mice
- Fed control, Western (40% fat), or Ketogenic (80% fat) diets for 1 month
- Treatment with γ-secretase modulators and anti-inflammatory drugs (NSAIDs) for 6 months
- Measured effect on Aβ, oxidative stress, motor function, etc.

Results: No significant effects

Plasma leptin negatively correlates with brain Aβ

How does a plasma hormone regulate Aβ accumulation in the brain?
Leptin Signaling

Leptin

β-Secretase

γ-Secretase

JAK (Janus Kinase)

STAT (Signal Transducer and Activator of Transcription)

Nucleus

Decreased Appetite

Increased Energy Expenditure

Leptin Controls γ-Secretase Expression

Leptin Controls γ-Secretase Expression

A. B. C. D. E.

Non Diabetic, Normal Leptin

Non Diabetic, Low Leptin

Type 2 Diabetic

Leptin

BRAKE

Aβ

Pathology

Healthy Brain

Alzheimer’s Disease

Alzheimer’s Disease
Chronically Elevated Leptin Signaling

Leptin → Leptin Receptor → JAK (Janus Kinase) → STAT (Signal Transducer and Activator of Transcription)

Increased Appetite
Decreased Energy Expenditure
Increased Obesity Risk
Increased Diabetes Risk

Mouse Model of Leptin Resistance Induced Obesity and Type 2 Diabetes Mellitus (T2DM)

4 main genotypes:
- wild-type (WT)
- diabetic (db)
- Alzheimer’s disease (AD)
- diabetic/AD (db/AD)

db/AD Mice
**db/AD Mice**

A. GTT

B. Phenotype

C. IGF1

D. Gene type

---

**db/AD Mice Have Memory Problems**

A. Acquisition Trial Block (day)

B. Time to Target (s) (mean ± SEM)

---

**Analysis of Amyloid, Tau and PS1 Expression**

A. Protein (pg/mL)

B. PS1 (pg/mL)

C. PS1 (pg/mL)

D. PS1 (pg/mL)
Our simplistic ideas about amyloid were wrong! What will we do?

Vascular Corrosion Casting

http://www.vasqtec.com/

WT Mice
AD Mice

Complex Saccular Aneurism

db/AD Mice Have Vascular Pathology in the Brain

AD Mice
Complex Saccular Aneurism
db/AD Mice Have Vascular Pathology in the Brain

db/AD Mice Have Strokes

Prussian Blue + Neutral Red

db/AD Mice Have Strokes
Why do we care?

- Both diabetes and AD are current, and future, public health crises
  - AD prevalence is increasing
  - Diabetes epidemic
  - Confluence of AD and diabetes not well studied and may require unique treatment approaches
- No approved treatment for vascular dementia
  - Unique model of diabetes-related mixed dementia
    - Prominent vascular pathology
    - Use to test different treatment approaches

Therapy?

- What happens if we remove the Aβ? Does the vascular pathology get better? Worse?
- What about fixing leptin signaling?
- What about the tau pathology? Does it matter for VCID?
- What happens if we lower blood sugar
- What happens if we treat their blood pressure, even if they aren’t hypertensive?
- Can this lead to new drug targets?
  … let’s cover some examples

What about Blocking Aβ?

- Does removal of Aβ ameliorate vascular pathology?
  - Aβ immunotherapy
  - γ-Secretase inhibition

Answer: targeting Aβ does not seem to be the way to go
How About Repairing Leptin in the Brain?

Diabetic Mouse

Faulty Brake

Brake Repaired?

AAV +

AAV -

Yup, we can do this…

But: too impractical for clinical use?

What about Tau?

Adapted from: Rubinsztein & Mucke, Science, 314: 781-784, 2006
Is Tau Related to Leptin as well?

A. Tau Phosphorylation (pThr231)

B. Tau Phosphorylation (pSer202+pThr205)

Obese Mice WILL get tangles faster (AAV-P301L Tau)
But even with more tangles, they don't seem any worse...

So, is tau not important for cognitive dysfunction?

Okay... how about just lowering blood sugar?

- Lifestyle modifications
  - Diet
  - Exercise (Wash U NIDDK)
- Treat the diabetes?
  - Metformin (Diabetes Complications Consortium)

Answer: probably made them a teeny bit better, but pretty modest...

This is Making Me sad...
Didn’t you mention blood pressure?

- Angiotensin receptor antagonist
  - Telmisartan (Bright Focus Foundation)
  - Administered for ~6 months at doses too low to affect BP
  - Some indication that it improves vascular health

What about other targets?

Na⁺/H⁺ Exchanger 1 (NHE1)

Interesting, but why should anyone care? Well...
NHE1 is important in Stroke

What does NHE1 Do?

Amiloride is an Excellent Drug
Summary

• Removing $A_i$ doesn’t help, and may actually make things worse (by leading to more severe strokes)
• It looks possible to fix leptin signaling, but this may be impractical in humans; however, leptin itself has some promise as an agent to treat cognitive dysfunction
• Although tau may play a role in AD, it seems unlikely to be important in VCID (so far)
• Lowering blood sugar seems to mainly affect the AD related pathology, but is nonetheless somewhat promising because it’s simple (although effects small)
• Treating blood pressure, even in the absence of hypertension, is extremely promising
• Other targets, such as $Slc9a1$, look promising as future goals to pursue with some therapeutic potential