“New Hope: Stem Cell Therapy in Alzheimer's Disease”

Mathew Blurton-Jones, Ph.D.

Department of Neurobiology & Behavior
Institute for Memory Impairments and Neurological Disorders
Sue and Bill Gross Stem Cell Research Center
University of California, Irvine
A NEW HOPE:

*Stem Cell Therapy in Alzheimer's Disease*
• What are stem cells and what types of stem cells might be used for Alzheimer’s disease research or therapies?

• Examining the effects of Neural Stem Cell transplantation in a transgenic mouse model of AD.

• What are some of the key challenges to clinical translation?

• How might induced pluripotent stem cell help us to understand or treat AD?
What is a Stem Cell?

Stem cells can make precise copies of themselves over and over again and stem cells can “differentiate” or mature into several cell types.
What are Stem Cells?

the raw material from which a body is built

**Pluripotent Stem Cells**
- In vivo fertilized egg
- 8 cell embryo
- Cultured undifferentiated stem cells
- Neural cells
- Cardiac muscle
- Blood cells

**Multipotent Stem Cells**
- Brain
- Neural cells
- Cardiac muscle
- Blood cells

40+ years of life saving treatments
Alzheimer Disease (AD)

- Could stem cells be used to replace the neurons that have been lost in AD?

It is very unlikely that a cell-replacement approach could be developed to treat advanced AD
For a **Cell Replacement** therapy you would need to:

1) Make appropriate neural stem cells in vitro

2) Transplant cells that can survive and differentiate into multiple different types of mature neurons

3) Have the transplanted neurons extend axons and dendrites into appropriate target regions.

4) Form appropriate synaptic connections
This is likely why so few studies have examined Stem Cells transplantation for AD
So why did we bother studying Stem Cell Transplantation for AD?

**Behavioral improvement in a primate Parkinson’s model is associated with multiple homeostatic effects of human neural stem cells**

D. Eugene Redmond, Jr.*1‡, Kimberly B. Bjugstad3, Yang D. Teng5, Vaclav Ourednik7, Jitka Ourednik8, Dustin R. Wakeman**9, Xuejun H. Parsons**3, Rodolfo Gonzalez**3*, Barbara C. Blankard1, Seung U. Kim**,*


**Human neural progenitors deliver glial cell line-derived neurotrophic factor to parkinsonian rodents and aged primates**

S Behrstock1*, A Ebert1, J McHugh1, S Vosberg2, J Moore1, B Schneider1, E Capowski1, D Heif3, J Kordower1, P Aebischer* and CN Svenisen1

**Multimodal Actions of Neural Stem Cells in a Mouse Model of ALS: A Meta-Analysis**

Yang D. Teng et al.

*Sci Transl Med* 4, 165ra164 (2012);

Examination of mesenchymal stem cell-mediated RNAi transfer to Huntington's disease affected neuronal cells for reduction of huntingtin

Scott D. Olson, Amal Kambal, Kari Pollock, Gaela-Marie Mitchell, Heather Stewart, Stefanos Kalomoiris, Whitney Cary, Catherine Nacey, Karen Pepper, Jan A. Nolta *
Can Neural Stem Cell Transplantation Affect Pathology or Cognition in a Mouse Model of AD?
Mouse Neural Stem Cells (NSCs)

- Neurons
- Astrocytes
- Oligodendrocytes
Why did we choose Green mice instead of Red ones?
AD Neuropathology

- **Amyloid Plaques**: β-amyloid peptide (Aβ)
- **Neurofibrillary Tangles**: hyperphosphorylated tau
- **Neuronal and Synaptic loss**: Aβ oligomers

**Synapse loss is the best correlate to dementia**
Triple Transgenic Mice (3xTg-AD) Exhibit Age-Dependent Accumulation of Plaques and Tangles
Transplantation of mNSCs into 18-month old 3xTg-AD mice
3xTg-AD mice exhibit deficits in spatial memory in the Morris Water Maze
Neural Stem Cells improve learning in Aged AD Transgenic Mice

Blurton-Jones et al., PNAS, 2009
Neural Stem Cells Improve Memory in Aged AD Transgenic Mice

Blurton-Jones et al., PNAS, 2009
How does NSC transplantation improve cognition in 3xTg-AD mice?
NSCs engraft into the host hippocampus

Blurton-Jones et al., PNAS, 2009
Most NSCs differentiate into astrocytes or oligodendrocytes
Only a small number of GFP+ neurons are detected

Blurton-Jones et al., PNAS, 2009
How does NSC transplantation improve cognition in 3xTg-AD mice?

-Probably not by making new neurons
Do NSCs effect AD Pathology?
NSC transplantation has no effect on Aβ pathology

Blurton-Jones et al., PNAS, 2009

Bar graphs showing soluble and insoluble Aβ levels in NSC transplantation compared to vehicle control, with images of Aβ fibrils and oligomers.
NSC transplantation also has no effect on tau pathology

Blurton-Jones et al., PNAS, 2009
How does NSC transplantation Improve Cognition in 3xTg-AD mice?

-not by altering AD pathology
**Synapse loss** is the best correlate to the severity of dementia in AD

Synapses in a culture dish: little red dots

Synapses in the brain: 1 quadrillion ($10^{15}$) little red dots

NSCs increase Synaptic Density within the hippocampus

Blurton-Jones et al., PNAS, 2009
Neurotrophins are a group of proteins that promote growth and synapse formation.

Neurotrophin levels are decreased in AD and PD.
NSCs elevate levels of Brain-Derived Neurotrophic Factor (BDNF) within the brain

Blurton-Jones et al., PNAS, 2009
NSCs BDNF Synapses Cognition
Is NSC-derived BDNF necessary for improved memory?

Stable knockdown of BDNF in NSCs:
Repeat the Experiment:

BDNF-depleted NSCs fail to improve cognition and have a diminished effect on synaptic density.

Blurton-Jones et al., PNAS, 2009
Recent studies have now also found similar improvements in cognition with Stem Cell Transplantation.

Table 1: Studies investigating transplantation of stem/progenitor cells for neurocognitive enhancement in Alzheimer’s disease

<table>
<thead>
<tr>
<th>Investigator</th>
<th>Animal model</th>
<th>Impaired animals transplanted with cells</th>
<th>Injected site</th>
<th>Craft</th>
<th>Cells injected per site</th>
<th>Behavioral test used</th>
<th>Time after injection</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Li et al. [60]</td>
<td>Bilateral Aβ injection into Hp</td>
<td>7</td>
<td>Hp</td>
<td>BMSC, BMSC-NGF, normal saline</td>
<td>200,000-300,000, 2 sites</td>
<td>MWM</td>
<td>8 days</td>
<td>Improvement with BMSC and BMSC-NGF</td>
</tr>
<tr>
<td>Lee et al. [70]</td>
<td>APP/PS1 double heterozygous mutant mice</td>
<td>15</td>
<td>Hp</td>
<td>BM-MSCs, NIH 3T3 as control</td>
<td>10,000,2 sites, biweekly</td>
<td>MWM</td>
<td>Approx. 1 month after first transplantation</td>
<td>BM-MSCs transplanted animals perform better than control transplanted rat</td>
</tr>
<tr>
<td>Tang et al. [54]</td>
<td>Bilateral Aβ injection into Hp</td>
<td>6</td>
<td>Hp</td>
<td>NPCs differentiated from mouse ES cells</td>
<td>1,000,000, 2 sites</td>
<td>MWM</td>
<td>4 weeks and 16 weeks</td>
<td>Rats grafted with NPCs had improved spatial learning</td>
</tr>
<tr>
<td>Moghadam et al. [57]</td>
<td>Ibotenic acid lesion of NBM</td>
<td>6</td>
<td>NBM</td>
<td>NPCs, FNPCs, ESCs, vehicle</td>
<td>200,000, 1 site</td>
<td>MWM</td>
<td>4 weeks</td>
<td>NPCs and FNPCs enhanced learning, ESCs impaired learning and formed tumors</td>
</tr>
<tr>
<td>Wang et al. [59]</td>
<td>Ibotenic acid lesion of NBM</td>
<td>10 (NPCs), 6 (ESCs)</td>
<td>Frontal association cortex and barrel field of S1</td>
<td>NPCs, ESCs, vehicle</td>
<td>4,000-20,000, 4 sites</td>
<td>8-arm radial maze</td>
<td>8 weeks</td>
<td>NPCs enhanced learning, ESCs impaired learning and formed tumors</td>
</tr>
<tr>
<td>Yamasaki et al. [50]</td>
<td>Transgenic model of neuronal injury</td>
<td>8 to 13</td>
<td>Hp</td>
<td>NSCs, vials</td>
<td>100,000, 2 sites</td>
<td>Object recognition, place recognition</td>
<td>1 month and 3 months</td>
<td>NSCs significantly enhanced memory after 3 months, but not after 1 month</td>
</tr>
<tr>
<td>Xu et al. [55]</td>
<td>Fimbria-fornix lesion</td>
<td>8</td>
<td>Ventricle</td>
<td>NSCs, NSCs with BDNF</td>
<td>50,000, 1 site, biweekly</td>
<td>Y-maze</td>
<td>4 weeks</td>
<td>NSCs and BDNF injections enhanced learning and memory</td>
</tr>
<tr>
<td>Xu et al. [56]</td>
<td>Fimbria-fornix lesion</td>
<td>8</td>
<td>Basal forebrain</td>
<td>NSCs, glia</td>
<td>50,000, 1 site</td>
<td>Y-maze</td>
<td>4 weeks</td>
<td>NSCs enhance learning and memory more than glia</td>
</tr>
<tr>
<td>Burlton-Jones et al. [53]</td>
<td>3xTg-AD mice</td>
<td>18</td>
<td>Hp</td>
<td>NSCs, vehicle</td>
<td>100,000, 2 sites</td>
<td>MWM, context-dependent novel object recognition</td>
<td>1 month</td>
<td>BDNF is essential for NSC-induced cognitive rescue</td>
</tr>
</tbody>
</table>
What are some of the key challenges to clinical translation?

- Human NSCs
- Longer-term studies
Can we translate this towards a clinical application?

- Requires clinical grade, GMP Human NSCs
- Requires testing human cells in AD mouse models: Xenotransplantation
GMP-compliant Human Neural Stem Cells

- Cryopreserved cell banks
- Allogeneic, unmodified cell
  - No pre-differentiation
- Completed 2 Phase I Safety Studies
  - Neuronal Ceroid Lipofuscinosis
  - Pelizaeus-Merzbacher Disease
- 2 ongoing Phase I/II Studies
  - Spinal Cord Injury
  - Age-Related Macular Degeneration
ACHIEVING STABLE HUMAN STEM CELL ENGRAFTMENT AND SURVIVAL IN THE CNS:

Types of transplantation, from autograft, to isograft, to allograft, to xenograft. As discordance between donor and host increases, so does the risk of rejection.

Anderson et al., 2011
Regen Med 6:367-406
Can Transplantation of Human NSCs improve memory in immunosuppressed 3xTg-AD mice?

All mice immunosuppressed with: rCTLA-Ig, anti-CD40L, anti-LFA-1
hNSCs improve Memory in immunosuppressed 3xTg-AD mice

Both 3xTg-AD groups immunosuppressed with: rCTLA-Ig, anti-CD40L, anti-LFA-1
hNSCs improve Memory in immunosuppressed 3xTg-AD mice

Both 3xTg-AD groups immunosuppressed with: rCTLA-Ig, anti-CD40L, anti-LFA-1
Engraftment of Human NSCs into 20-month old 3xTg-AD mice

Red: Human cytoplasm (SC121)

Nestin
Doublecortin
Doublecortin
hGFAP

immunosuppression: anti-CD40L, rCTLA-Ig, anti-LFA-1
What are some of the key challenges to clinical translation?

- Human NSCs
- Longer-term studies
How can we study long-term Human Stem cell engraftment?

Lack T-cells, B-cells, and NK-Cells, yet have normal lifespans

Intraneuronal β-Amyloid Aggregates, Neurodegeneration, and Neuron Loss in Transgenic Mice with Five Familial Alzheimer’s Disease Mutations: Potential Factors in Amyloid Plaque Formation

- Highly aggressive plaque pathology
- Progressive Cell loss beginning at 9 months
Spleen flow cytometry confirms deletion of B, T, and NK lineages in Rag-5x mice

With Jason Weinger and Tom Lane
Rag2ko/il2rgko-5xfAD mice develop extensive Aβ pathology and gliosis

Sam Marsh
6-months after transplantation HuCNS-SC have survived and migrated extensively in 5x-Rag mice.
So is there a “Dark Side” to stem cell therapies?

Snake Oil salesman with beautiful websites and miraculous promises have quickly jumped on the scene!
An unproven stem cell treatment in Russia caused brain tumors in a young boy

Stem cell therapy triggers tumor

A neural stem cell transplant from fetal cells performed in Russia led to a brain tumor in a teenage boy, researchers in this week’s PLoS Medicine report, raising concerns about the safety of neural stem cells treatments. MRI of brain lesion, courtesy of PLoS Medicine. The researchers confirmed that the cancer originated from the donor tissue, not the boy’s own cells. This is the first report of cancer following fetal neural stem cell transplant. However, outside experts raised concerns about th

By Tia Ghose | February 18, 2009

Donor-Derived Brain Tumor Following Neural Stem Cell Transplantation in an Ataxia Telangiectasia Patient

Presence of cells with no Y chromosome in the Boy’s tumor
The International Society for Stem Cell Research has created a campaign to inform people about the risks of “Stem Cell Tourism”.

Find out what's possible. Know what to ask.

We have all heard about the extraordinary promise that stem cell research holds for the treatment of human diseases. Clinics all over the world claim to offer stem cell treatments for a wide variety of conditions. But are all of these treatments likely to be safe and effective?

The ISSCR provides information to help you evaluate these claims. Learn more about what this site can provide.

View CBS' 60 Minutes (US) 2010 segment, "21st Century Snakeoil"

http://www.closerlookatstemcells.org/
Are there any other ways that stem cells could be used to understand or treat AD?
Induction of Pluripotent Stem Cells from Adult Human Fibroblasts by Defined Factors

Kazutoshi Takahashi,1 Koji Tanabe,1 Mari Ohnuki,1 Megumi Harita,1,2 Tomoko Ichisaka,1,2 Kiichiro Tomoda,3 and Shinya Yamanaka1,2,3,4,*

1Department of Stem Cell Biology, Institute for Frontier Medical Sciences, Kyoto University, Kyoto 606-8507, Japan
2CREST, Japan Science and Technology Agency, Kawaguchi 332-0012, Japan
3Gladstone Institute of Cardiovascular Disease, San Francisco, CA 94158, USA
4Institute for Integrated Cell-Material Sciences, Kyoto University, Kyoto 606-8507, Japan

*Correspondence: yamanaka@frontier.kyoto-u.ac.jp
DOI 10.1016/j.cell.2007.11.019

“Induced”

Pluripotent Stem Cells

Patient skin cells

+ 4 key genes
Can induced pluripotent stem cells (iPSC) be used to examine the molecular mechanisms that cause late-onset AD?

Late-onset AD is ~80% genetic.

Do multiple small genetic differences add up to shift the function or phenotype of brain cells?

Once these key differences are identified, iPSCs could also be used to screen for and test new drugs.
The UCI Alzheimer’s disease research center has begun generating a bank of patient-derived iPSCs to ask these questions.
Summary

• Neural stem cell transplantation improves cognition in a mouse model of Alzheimer’s disease.

• Understanding the mechanism by which NSCs influence cognition could identify additional therapeutic approaches.

• Translating findings toward the clinic will take some time and we need to be cautious about unregulated ‘clinics’ promising stem cell cures.

• Induced pluripotent stem cells could provide researchers with a whole new way to study and understand what drives AD and how we might treat it.
Acknowledgements

UC Irvine
Frank LaFerla
Rahasson Ager
Samuel Marsh
Joy Nerhus
Andy Agazaryan
Masashi Kitizawa
Kim Green
Wayne Poon
Thomas Lane
Jason Weinger
Wesley Chen
Natalie Sashkin Goldberg

UC San Diego
Eliezer Masliah
Brian Spencer

StemCells Inc.
Stephen Huhn
Alexandra Capela

Supported by:

National Institute on Aging

alzheimer's association