Profiling the neuroinflammatory response in AD — potential for personalized medicine and therapeutic modulation

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Microglial cells surround plaques and tangles
An autotoxic loop

- Initial insult
- Some neuronal death
- Deposition of debris
- Activation of microglia (with secretion of toxic materials)
  - Reinforcing cross-stimulation
- Activation of complement cascade (including formation of MAC)
- More rapid neuronal death

Adapted from McGeer and McGeer, Exp Gerontol 33:371-378
Inflammation has a purpose

Potentially damaging proteins –
Purpose – to clear debris
to destroy invading cells

Non-damaging proteins –
Purpose – to repair a wound
to decrease damaging proteins

Level

Time

Injury

Resolution
We can identify types of neuroinflammation

Macrophage

OR

Microglia
Determining the inflammatory state of the human Alzheimer’s disease brain

- Many postmortem tissue have used late-stage AD brain samples.

- **We hypothesized that there may be differences in the neuroinflammatory state as AD progresses.**

- We obtained tissue samples from early AD, late AD and age-matched, non-demented controls.

- Gene expression levels of neuroinflammatory markers measured by qRT-PCR from frozen brain tissue frontal cortex and cerebellum.

- Serum analyzed for predictive markers of the brain neuroinflammatory state.

- Clinical histories of patients examined to determine common non-AD factors that account for differences in the neuroinflammatory states.
## Human AD tissue characteristics

<table>
<thead>
<tr>
<th>Group</th>
<th>Age range (yr)</th>
<th>MMSE</th>
<th>ApoE4 status</th>
<th>Braak stage</th>
<th>PMI (hr)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Early AD</td>
<td>77-100 (mean = 87.2)</td>
<td>20-24</td>
<td>-/4 = 12, 4/4 = 3</td>
<td>3-5</td>
<td>1.0-6.5 (mean = 3.6)</td>
</tr>
<tr>
<td>Late AD</td>
<td>74-88 (mean 85.6)</td>
<td>0-13</td>
<td>-/4 = 6, 4/4 = 4</td>
<td>6</td>
<td>1.75-11.0 (mean = 5.4)</td>
</tr>
<tr>
<td>Control</td>
<td>77-96 (mean = 84.3)</td>
<td>28-30</td>
<td>-/4 = 10, 4/4 = 3</td>
<td>0-2</td>
<td>1.75-8.0 (mean = 4.2)</td>
</tr>
</tbody>
</table>
Cluster analysis revealed two distinct populations in the early AD group
Early AD brain is biased to either the M1 or M2a phenotype in the frontal cortex.
In the cerebellum, the inflammatory polarization does not exist.
End-stage AD brain does not show the same heterogeneity observed in early stage.
Next step....

• **Hypothesis**: The polarization of the neuroinflammatory state of the early AD brain to either M1 or M2 will significantly influence response to therapy.

• Serum from our samples were run on the Myriad-RBM human inflammation MAP.

• Our goal was to identify serum biomarkers predictive of the brain neuroinflammatory state.
Serum markers are predictive of brain neuroinflammatory state
Cerebrovascular disease risk factors are present with an M2 polarization

- We retrieved the following history:
  - Congestive heart failure
  - Angina
  - Hypertension
  - Peripheral vascular disease
  - Atrial fibrillation
  - Coronary artery bypass graft
  - Angioplasty

- We added up the number of risk factors present and analyzed this based on the brain neuroinflammatory profile.

- We found that the M2 phenotype is associated with the increased presence of cerebrovascular disease risk factors.
Can we modulate the inflammatory response and influence AD pathology?
Modulation to M2b

Macrophage

M1
- IL-1β
- TNFα
- IL-12
- IL-6
- LOW IL-10

M2a
- AG1
- MRC1
- YM1
- FIZZ
- IL-1Ra
- LOW IL-12

M2b
- CD86
- IL-10
- TNFα
- IL-1β
- IL-6
- LOW IL-12

M2c
- TGFβ
- IL-10
- MRC1
- Sphk1
- LOW IL-12

Immune complexes
Study design

• 120 seven month old APP/PS1 mice were assigned to one of 4 treatment groups.
  – Saline.
  – Anti-Aβ antibody (6E10, Aβ3-8, IgG1)
  – IVIg (composed primarily of IgG1 and IgG2)
  – Mouse IgG (composed primarily of IgG1 and IgG2a).

• Intracranial injections were performed bilaterally in the frontal cortex and hippocampus.

• The right side was dissected for biochemistry and the left was fixed and processed for histology.
Aβ is reduced by all treatments but anti-Aβ antibody shows a more rapid reduction.
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Microglial activation is induced by all three antibody injections
Microglial activation peaks later with IVIg than anti-Aβ antibody.
Neuroinflammatory phenotypes are modulated by antibody presence in the brain.
Summary and conclusions

• Neuroinflammatory profiles can be used to phenotype the inflammatory response in the brain.

• Early AD brain exhibits diverse neuroinflammatory phenotypes that will likely directly influence response to therapeutic interventions.

• M2 inflammatory phenotypes in the brain are associated with elevated cerebrovascular disease risk factors.

• IVIg, when administered intracranially, promotes an M2b phenotype that appears to precede the amyloid reductions.

• We hypothesize that “modulation” of the neuroinflammatory phenotype is a potential therapeutic approach for the treatment of Alzheimer’s disease.
Our working hypotheses

• Neuroinflammatory phenotypes can directly influence Alzheimer’s disease pathology onset and progression.

• The co-morbidity of vascular dementia with Alzheimer’s disease will require different therapeutic approaches than pure AD alone.
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