ADNI Biomarker Core Report

Leslie M Shaw & John Q Trojanowski

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Michal Figurski    Leona Fields
Teresa Waligorska    Sarah Pan
ADNI Biomarker core 2013- July 2014

• Studies reported in the ADNI/LONI website & studies newly approved by RARC/NIA/ADNI using ADNI biofluids
• 2014 ADNI II batch analyses of CSF completed, uploaded on ADNI/LONI website June 2014
• mrm/tandem mass spectrometry reference method for Aβ1-42, progress report
• Planned analyses
ADNI Biomarker core 2013-July 2014:
Add-on studies reported on the ADNI/LONI website & add-on studies newly approved by RARC/NIA/ADNI using ADNI biofluids

Reported

- mrn/tandem mass spectrometry of 567 tryptic peptides associated with 221 proteins, Caprion/FNIH/ADNI/PPSB in ADNI I CSF

Approved/shipped or to be shipped

- YKL40 & Vilip1 in longitudinal ADNI CSF, WashU, Anne Fagan, 4th qtr 2013
- DDE* in ADNI serum, EmoryU, Allan Levey, 2nd qtr 2014
- Neurogranin in BASELINE CSF of ADNI I subjects, UGothenberg, Sweden, Kaj Blennow, 2nd qtr 2014
- Tau in plasma of ADNI I BASELINE subjects, UGothenberg, Sweden, Kaj Blennow, 3rd qtr 2014
- Phosphorylated α-synuclein in ADNI I BL CSF, UWash, Jing Zhang, 2nd qtr 2014

*DDE-a major breakdown product of the insecticide DDT that is highly fat soluble, very long biological half-life, potential risk factor for neurodegeneration. JAMA Neurology 2014.
Aliquot counts by study subject

- Updated as of 4/14 2014 on the ADNI/LONI website
- Aliquot counts for plasma, serum, CSF, urine
- Total volume and # of pristine aliquots available
- Aliquots utilized for Biomarker Core measurements
- Aliquots sent to investigators who had RARC/NIA/ADNI-approved studies
2014 batch analyses of ADNI II CSFs

- N=428 ADNI II CSFs + 17 pristine randomly selected replicates assayed using the AlzBio3 RUO immunoassay
- 275 BASELINE samples; 153 have a paired BL(2013) + 24 month sample
- Results uploaded on ADNI/LONI website June 2014.
- QC data summary in next ppt.
# 2014 batch analyses of ADNI II CSFs

<table>
<thead>
<tr>
<th></th>
<th>Sample</th>
<th>N</th>
<th>Mean ± SD</th>
<th></th>
<th>N</th>
<th>Mean ± SD</th>
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<td><strong>2013</strong></td>
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<td><strong>2014</strong></td>
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<tr>
<td><strong>t-tau</strong></td>
<td>CSF abnormal pool #53</td>
<td>25</td>
<td>128 ± 10.4</td>
<td><strong>t-tau</strong></td>
<td>14</td>
<td>120 ± 4.9</td>
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<td></td>
<td>CSF normal pool #54</td>
<td>25</td>
<td>65.9 ± 6.0</td>
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<td>14</td>
<td>59.8 ± 4.7</td>
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<td><strong>Aβ_{1-42}</strong></td>
<td>CSF abnormal pool #53</td>
<td>25</td>
<td>148 ± 9.4</td>
<td><strong>Aβ_{1-42}</strong></td>
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<td>148 ± 8.4</td>
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<tr>
<td></td>
<td>CSF normal pool #54</td>
<td>25</td>
<td>236 ± 18</td>
<td></td>
<td>14</td>
<td>249 ± 13.7</td>
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<tr>
<td><strong>p-tau_{181}</strong></td>
<td>CSF abnormal pool #53</td>
<td>25</td>
<td>26.5 ± 1.4</td>
<td><strong>p-tau_{181}</strong></td>
<td>14</td>
<td>28.0 ± 1.7</td>
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<tr>
<td></td>
<td>CSF normal pool #54</td>
<td>25</td>
<td>19.1 ± 1.2</td>
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<td>14</td>
<td>20.4 ± 1.3</td>
</tr>
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</table>

## Test-retest performance
- **Linear regression R^2 values**
  - Tau, 0.986
  - Aβ_{42}, 0.924
  - ptau_{181}, 0.988
- **Bland-Altman avg %CV**
  - Tau, 7.2%
  - Aβ_{42}, 5.6%
  - ptau_{181}, 4.9%
<table>
<thead>
<tr>
<th></th>
<th>N</th>
<th>% Apoe ε4</th>
<th>t-tau (pg/mL)</th>
<th>Aβ1-42 (pg/mL)</th>
<th>p-tau181 (pg/mL)</th>
<th>t-tau/Αβ1-42</th>
<th>p-tau181/Αβ1-42</th>
<th>LRTAA2i</th>
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<tbody>
<tr>
<td></td>
<td>AD</td>
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<tr>
<td>ADNI I</td>
<td>100</td>
<td>69.0</td>
<td>122±58</td>
<td>144±41</td>
<td>42±20</td>
<td>0.92±0.48</td>
<td>0.32±0.19</td>
<td>0.79±0.29</td>
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<td>ADNI II</td>
<td>131</td>
<td>66.4</td>
<td>133±62</td>
<td>132±33</td>
<td>56±26</td>
<td>1.1±0.61</td>
<td>0.46±0.28</td>
<td>0.63±0.38</td>
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<td>LMCI</td>
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<tr>
<td>ADNI I</td>
<td>196</td>
<td>54.1</td>
<td>103±61</td>
<td>164±55</td>
<td>36±18</td>
<td>0.75±0.62</td>
<td>0.26±0.18</td>
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<td>ADNI II</td>
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<td>57.4</td>
<td>101±56</td>
<td>158±49</td>
<td>49±28</td>
<td>0.72±0.48</td>
<td>0.35±0.24</td>
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<td>ADNI II</td>
<td>273</td>
<td>42.1</td>
<td>77±49</td>
<td>184±51</td>
<td>37±21</td>
<td>0.49±0.46</td>
<td>0.24±0.19</td>
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<td>ADNI I</td>
<td>114</td>
<td>23.7</td>
<td>70±30</td>
<td>206±55</td>
<td>25±15</td>
<td>0.39±0.27</td>
<td>0.14±0.13</td>
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<td>ADNI II</td>
<td>157</td>
<td>26.7</td>
<td>68±34</td>
<td>196±51</td>
<td>35±19</td>
<td>0.39±0.27</td>
<td>0.20±0.16</td>
<td>0.32±0.34</td>
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<td>ADNI I</td>
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<tr>
<td>ADNI II</td>
<td>93</td>
<td>33.3</td>
<td>65±32</td>
<td>201±49</td>
<td>38±21</td>
<td>0.36±0.24</td>
<td>0.21±0.17</td>
<td>0.31±0.32</td>
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</table>
CSF biomarkers for ADNI GO+2 subjects stratified by #APOE e4 alleles

- Tau mean (pg/mL)
- Abeta1-42 mean (pg/mL)
- p-tau181 mean (pg/mL)

<table>
<thead>
<tr>
<th>Diagnostic Groups</th>
<th>NC</th>
<th>EMCI</th>
<th>LMCI</th>
<th>AD</th>
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</thead>
<tbody>
<tr>
<td>0 alleles (n)</td>
<td>44</td>
<td>158</td>
<td>66</td>
<td>177</td>
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<td>1 allele (n)</td>
<td>61</td>
<td>96</td>
<td>63</td>
<td>66</td>
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<tr>
<td>2 alleles (n)</td>
<td>26</td>
<td>19</td>
<td>26</td>
<td>7</td>
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</table>

- Standard deviation

see text below for stats
ADNI Longitudinal Biomarkers Changes

Adni 1

Aβ42

Toledo et al, Acta Neuropath, 2013
ADNI 2: Biomarker Core
mrm LC/MSMS method for Aβ peptides in CSF

- Further improved using a Xevo TQ-S tandem mass spectrometer; CSF aliquot size 100 µL
- Accuracy-based measurement of Aβ_{1-42}, Aβ_{1-40} & Aβ_{1-38} for all ADNI CSF samples
- Round Robin study completed (4 centers) in collaboration with Kaj Blennow, manuscript submitted
- Pilot assessment of IRMM-prepared Aβ_{1-42} is underway
- Will help answer questions about potential contribution of other metabolites, eg, Aβ_{1-40}, Aβ_{1-38} to utility of Aβ_{1-42} and other APP species, tau fragments.
Candidate reference methods for CSF Aβ42 published

Mass Spectrometry-Based Candidate Reference Measurement Procedure for Quantification of Amyloid-β in Cerebrospinal Fluid

Andreas Leinertbach,1† Josef Fanni,2,† Thomas Döllner,1 Andreas Huber,1 Tobias Bittner,1 Ulf Andreae,2 Johan Gobom,2 Henrik Zetterberg,2,3 Uwe Kobold,† Erik Portellus,2 and Kai Blennow2† on behalf of the IFCC Scientific Division Working Group on CSF proteins

Qualification of a Surrogate Matrix-Based Absolute Quantification Method for Amyloid-β_{42} in Human Cerebrospinal Fluid Using 2D UPLC-Tandem Mass Spectrometry

Magdalena Korecka4, Teresa Waligorska4, Michal Figurski4, Jon B. Toledo4,†, Steven E. Arnold4,†, Murray Grossman4, John Q. Trojanowski4,4 and Leslie M. Shaw4,†,*

Four laboratory Round Robin study on SRM Aβ42 methods
- 12 CSF samples
- On sample = Candidate Reference Material

SRM mass spec suitable as a Reference Measurement Procedure (RMP) for CSF Aβ 42
Support of standardization efforts

- ADNI-longterm commitment to standardization of all methods
- Alz Assn Global Biomarker Standardization Consortium
  - Analytical methods standardization--strong support for improved performance of existing and new immunoassays for CSF biomarkers
  - Support for mrm/tandem mass spectrometry for direct measurement of absolute $A\beta_{1-42}$ concentration
  - IFCC/IRMM project to develop reference $A\beta_{1-42}$ peptide material and using mrm/msms and large pools of CSF with accurately measured $A\beta_{1-42}$
  - Need same for t-tau
- CAMD(Coalition Against Major Diseases) has made a substantial commitment to support use of HV and CSF AD biomarkers in treatment trials
  - Hippocampal volume
  - CSF AD biomarkers
- Close collaboration with Japan ADNI on a joint effort to standardize lab-to-lab method performance of AlzBio3 immunoassay
  - Completed development and testing of a unified test procedure
  - A follow up study now underway for a larger scale assessment in AD and normal CSF samples
Planned Analyses of ADNI CSF Biomarker data
Leslie M. Shaw and John Q. Trojanowski

• Continue systematic documentation of the analytical performance of the AlzBio3 immunoassay and the validated mrm mass spectrometry assay for Aβ42/40/38
• Document the CSF biomarker characteristics in ADNI II subjects to test for replication of these findings in ADNI I
  • Bimodal distribution for Aβ1-42 and mixture modeling for cutpoint determination
  • Incidence of CSF biomarker based pathology across the ADNI subject groups
  • Predictive performance of BASELINE CSF biomarkers for decline in memory, cognition and daily function at 2 years
• Establish the clinical utilities of the qualified mrm/mass spectrometry method for Aβ1-42, Aβ1-40, Aβ1-30 and ratios of the latter two to Aβ1-42
• Assess the potential for new biomarkers to add sensitivity and specificity to CSF Aβ1-42, tau and p-tau181 for detection of AD neuropathology and add for prediction of progression from MCI to AD dementia, decline of cognition, memory and functions of daily living. This is an area we expect to expand on and build on for the ADNI III competitive renewal.
Planned Analyses of ADNI CSF Biomarker data

Longitudinal CSF biomarker data

Characterize the 2 yr trajectories in ADNI II BASELINE + 24 months for AD, LMCI, EMCI, SMC and NC. For those subject trajectories that are “normal” at BASELINE but move significantly toward “pathologic” and those that remain stable normal-test for progression of cog performance over time. Does the progression to “pathologic” CSF biomarker(s) predict or correlate with progression of cog perf over time; does the “stable normal” trajectory predict lack of cog/mem/functional decline?
A. **New biomarkers**{natural lead-in to planning analyses for ADNI III}.  

We expect that by the end of 2014 there will have been uploaded on the ADNI website sets of new biomarker data including CSF biomarkers and serum or plasma biomarkers, mostly in ADNI I study subjects. This affords us the opportunity to collaborate with others on the analyses of these data sets including:

1. **CSF**
   a. VILIP-1 & YKL40
   b. mrm/mass spectrometry (Caprion/FNIH/PPSB/ADNI) study protein/peptides profiles
   c. neurogranin
   d. $\text{A}_\beta_{1-42}$ oligomers (in collaboration between UPENN ADRC and Mary Savage) using (non-ADNI) UPENN CSF samples in a study that will be planned during summer, 2014.

2. **serum or plasma**
   a. DDE-a major breakdown product of the insecticide DDT that is highly fat soluble, very long biological half-life, potential risk factor for neurodegeneration. JAMA Neurology 2014
   b. t-tau in plasma

We are very interested to study the relationships of BASELINE values of these new CSF biomarkers to prediction of progression from MCI to AD dementia, and to decline of cognition, memory and functions of daily living and to CSF levels of $\text{A}_\beta_{1-42}$, t-tau and $p$-tau$_{181}$. 
It takes a great team effort!

John Q Trojanowski
Magdalena Korecka
Magdalena Brylska
Teresa Waligorska
Michal Figurski
Leona Fields
Sarah Pan
Virginia M-Y Lee
Chris Clark*
Steve Arnold
Hugo Vanderstichele

Margaret Knapik-Czajka
Ravi Patel
Pawel Zero
William Hu
Ju Hee Kang
Jon Toledo
Anne Fagan
Uwe Christians
Kaj Blennow
Henrik Zetterberg

Holly Soares
Adam Simon
Robert Dean
Eric Siemers
Piotr Lewczuk
William Potter
Rand Jenkins
Erin Chambers

*Deceased

Supported by the NIH/NIA and families of our patients

ADNI investigators include: (complete listing available at www.loni.ucla.edu\ADNI\Collaboration\ADNI_Manuscript_Citations.pdf).