Longitudinal PD Biomarker Studies: DeNoPa and PPMI

Prof. Dr. Brit Mollenhauer
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**CSF α-synuclein** in independent cross sectional studies and a cohort study

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**The Lancet Neurology**

α-Synuclein and tau concentrations in cerebrospinal fluid of patients presenting with parkinsonism: a cohort study

Ihr Mollenhauer, Joseph Lucatelli, Walter Schulz-Schaeffer, Frederike Siewel-Doring, Claudia Trenkwalder, Michael G. Schneider
Remaining Questions

• Are CSF total α-synuclein levels decreased in
  – early, de-novo PD
  – Premotor PD

• Are CSF total α-synuclein levels helpful to monitor disease progression?

• Are there any other (more accurate) biomarker, that can support the clinical diagnosis and monitor progression
Single-center  
National  
Observational  
159 Early, at enrolment drug naive PD subjects (UK Brain Bank)  

Multi-center (24 sites)  
International  
Observational  
423 Early, at enrolment drug naive PD subjects (UK Brain Bank/monosymptomatic tremor) PLUS Dopaminergic deficit  

110 matched (age, gender, education) healthy controls  
FOCUS: non-motor symptoms, fluid biomarker  

196 matched (age, gender) healthy controls  
FOCUS: functional and structural imaging, fluid biomarker  

Longitudinal  
Follow-up: every 24 months  
Autopsy verification of diagnoses ongoing  

Longitudinal  
Follow-up: every 6-12 months (V1/St visit)  
Autopsy verification of diagnoses planned  
Sharing of data and fluids (ppmi-info.org)  

EXPLORATION OF NEW MARKER  
VERIFICATION OF NEW MARKER
The long-term follow-up study of early (drug-naïve) Parkinson’s Disease patients and healthy controls for non-motor symptoms and biomarkers

Paracelsus-Elena Klinik, Kassel
Hospital for Movement Disorders
1937-2013

Linked to Göttingen University

German Register for Clinical trials (DRKS00000540) according to the WHO Trial Registration Data Set.
36 patients follow-up after 20 years

- 48% orthostatic Hypotension
- 74% Hallucinations and/or Depression
- 78% Dysarthria
- 48% Dysphagia
- 87% Falls (Bone fractures 35%)
- 81% Freezing
- 70% Daytime sleepiness
- 83% Dementia
The emergence of non-motor symptoms 10 years before the onset of PD
Data are mean (SD, range)  

<table>
<thead>
<tr>
<th></th>
<th>HC N=110</th>
<th>PD n=159</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>age (years)</strong></td>
<td>64.7 (6.83, 44.00-84.00)</td>
<td>65.3 (9.70, 40.00-85.00)</td>
<td>0.610</td>
</tr>
<tr>
<td><strong>sex</strong></td>
<td>female 67/ male 43</td>
<td>female 105/ male 54</td>
<td>0.811</td>
</tr>
<tr>
<td></td>
<td>(60/ 40)</td>
<td>(66/ 34)</td>
<td></td>
</tr>
<tr>
<td><strong>Number of comorbidities</strong></td>
<td>2.5 (1.53, 0-7)</td>
<td>2.3 (1.61, 0-8)</td>
<td>0.333</td>
</tr>
<tr>
<td><strong>disease duration [months]</strong></td>
<td>26.9 (37.11, 2-00-240.00)</td>
<td>26.9 (37.11, 2-00-240.00)</td>
<td></td>
</tr>
<tr>
<td><strong>Hoehn &amp; Yahr score</strong></td>
<td>1.8 (0.65, 1.00-3.00)</td>
<td>1.8 (0.65, 1.00-3.00)</td>
<td></td>
</tr>
<tr>
<td><strong>UPDRS total score</strong></td>
<td>29.2 (14.67, 4.00-68.00)</td>
<td>29.2 (14.67, 4.00-68.00)</td>
<td></td>
</tr>
<tr>
<td><strong>UPDRS motor score before levodopa</strong></td>
<td>n=158</td>
<td>n=158</td>
<td></td>
</tr>
<tr>
<td><strong>Tremor score $</strong></td>
<td>0.3 (0.34, 0.2-0.00)</td>
<td>0.3 (0.34, 0.2-0.00)</td>
<td></td>
</tr>
<tr>
<td><strong>PIGD Score $</strong></td>
<td>0.7 (0.58, 0-2.50)</td>
<td>0.7 (0.58, 0-2.50)</td>
<td></td>
</tr>
</tbody>
</table>

*DeNoPa-Cohort*  

Mollenhauer et al., 2013
Recruitment and first Follow-up

PD Patients

1. Baseline assessment (BL) - n=159
2. 2 year Follow-up assessment - n=147
3. Follow-up data analysis (24FU) - n=123
   - Drop-out: n=8 lost for 24FU, n=4 died before 24FU
   - Other neurological Disorders at 24FU (OND; n=24)*

Healthy Controls (HC)

1. Baseline assessment (BL) - n=110
2. 2 year Follow-up assessment - n=107
3. Follow-up data analysis (24FU) - n=106
   - Drop-out: n=1 lost for 24FU, n=2 died before 24FU
   - N=1 with PD like features at 24FU

* OND: MSA, PSP, DLB, Dystonia, unclear diagnosis
### Study population
- 400 de novo PD subjects ("possible PD" with positive DaT and unmedicated)
- 200 age- and gender-matched healthy controls
- Subjects will be followed for a minimum of 3 years and a maximum of 5 years

### Assessments / Clinical data collection
- Motor assessments
- Neuropsychiatric/cognitive testing
- Olfaction
- DaTSCAN imaging, MRI

### Biologic collection
- DNA collected at screening
- Serum and plasma collected at each visit; urine collected annually
- CSF collected at baseline, 6 months, 12 months and annually thereafter
- Samples aliquotted and stored in central biorepository

### PD treatment
- De novo for ~6 months
- Can participate in other clinical trials (including interventional trials) after 12 months

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**Recruitment finalized May 2013**

423 PD subjects
196 healthy controls (matched for age and gender)
64 SWEDD subjects

**ANALYSES ONGOING**
Original Investigation

Association of Cerebrospinal Fluid β-Amyloid 1-42, T-tau, P-tau\textsubscript{181}, and α-Synuclein Levels With Clinical Features of Drug-Naive Patients With Early Parkinson Disease

Ju-Hee Kang, MD; David J. Irwin, MD; Alice S. Chen-Plotkin, MD; Andrew Siderowf, MD; Chelsea Caspell, MS; Christopher S. Coffey, PhD; Teresa Waligorska, MS; Peggy Taylor, ScD; Sarah Pen, MPH; Mark Frasier, PhD; Kenneth Marek, MD; Karl Kieburtz, MD, MPH; Donna Jennings, MD; Tanya Simuni, MD; Caroline M. Tanner, MD, PhD; Andrew Singleton, PhD; Arthur W. Toga, PhD; Schini Chowdhury, MA; Brit Mollenhauer, MD, John O. Trojanowski, MD, PhD; Leslie M. Shaw, PhD; and the Parkinson’s ProgressionMarkers Initiative

Table 4. Comparison of Cerebrospinal Fluid Biomarkers Between Patients With Parkinson Disease Who Have the Tremor-Dominant vs Postural Instability–Gait Disturbance Motor Phenotype

<table>
<thead>
<tr>
<th>Biomarker</th>
<th>Mean (SD) PIGD-PD (n=14)</th>
<th>Mean (SD) TD-PD (n=43)</th>
<th>P Value for Mann-Whitney U Test\textsuperscript{a}</th>
<th>Mean (SD) HCs (n=39)</th>
<th>Mean (SD) IND-PD (n=6)</th>
<th>P Value for Kruskal-Wallis Test\textsuperscript{b}</th>
<th>Significance (PIGD-PD vs HC, After Dunn Test\textsuperscript{c})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aβ1-42, pg/mL</td>
<td>211.4 (45.0)</td>
<td>236.2 (46.8)</td>
<td>.03</td>
<td>242.8 (50.0)</td>
<td>215.5 (25.0)</td>
<td>.02</td>
<td>Yes, .01 &lt; P &lt; .05</td>
</tr>
<tr>
<td>T-tau, pg/mL</td>
<td>39.3 (28.27)</td>
<td>50.3 (24.01)</td>
<td>.05</td>
<td>53.9 (19.33)</td>
<td>31.2 (9.97)</td>
<td>.007</td>
<td>Yes, .01 &lt; P &lt; .05</td>
</tr>
<tr>
<td>P-tau\textsubscript{181}, pg/mL</td>
<td>18.0 (6.74)</td>
<td>22.5 (8.17)</td>
<td>.04</td>
<td>24.9 (8.45)</td>
<td>17.7 (4.97)</td>
<td>.005</td>
<td>Yes, .001 &lt; P &lt; .01</td>
</tr>
<tr>
<td>α-Syn, pg/mL</td>
<td>892.8 (542.4)</td>
<td>1185 (649.6)</td>
<td>.06</td>
<td>1264 (425.7)</td>
<td>782.6 (150.1)</td>
<td>.008</td>
<td>Yes, .01 &lt; P &lt; .05</td>
</tr>
<tr>
<td>Subjects with CSF Hib &gt;200 ng/mL</td>
<td>766.3 (446.3)</td>
<td>1122 (451.8)</td>
<td>.03</td>
<td>1267 (443.5)</td>
<td>775.9 (184.8)</td>
<td>.01</td>
<td>Yes, .001 &lt; P &lt; .01</td>
</tr>
<tr>
<td>T-tau/Aβ1-42 ratio</td>
<td>0.211 (0.213)</td>
<td>0.225 (0.145)</td>
<td>.11</td>
<td>0.240 (0.141)</td>
<td>0.151 (0.072)</td>
<td>.03</td>
<td>No</td>
</tr>
<tr>
<td>P-tau/Aβ1-42 ratio</td>
<td>0.093 (0.059)</td>
<td>0.104 (0.068)</td>
<td>.22</td>
<td>0.113 (0.075)</td>
<td>0.083 (0.026)</td>
<td>.25</td>
<td>No</td>
</tr>
<tr>
<td>P-tau/T-tau ratio</td>
<td>0.617 (0.398)</td>
<td>0.513 (0.217)</td>
<td>.76</td>
<td>0.491 (0.160)</td>
<td>0.588 (0.164)</td>
<td>.61</td>
<td>No</td>
</tr>
</tbody>
</table>
CSF BIOMARKERS REMAIN STABLE AT 1-YEAR FOLLOW-UP

**Analyte/Diagnosis** | Summary | Baseline (0 months) | 6 months | 12 months
--- | --- | --- | --- | ---
α-Syn PD | Mean (SE) | 412 | 128 | 166
CEU | N | 1974 (38.7) | 1838 (65.8) | 1861 (62.1)

**Analyte/Diagnosis** | Summary | Baseline (0 months) | 6 months | 12 months
--- | --- | --- | --- | ---
Aβ1-42 PD | Mean (SD) | 412 | 129 | 166
CEU | N | 370 (4.95) | 367 (8.57) | 376 (8.07)

α-Syn HC | Mean (SE) | 189 | 112 | 111
CEU | N | 2204 (79.2) | 2201 (86.7) | 2164 (91.1)

Aβ1-42 HC | Mean (SD) | 180 | 112 | 111
CEU | N | 377 (8.26) | 374 (9.35) | 389 (10.0)
## Assay development and validation for 3-5 relevant for PD progression aSyn species

**PI:** Brit Mollenhauer  
Peggy Taylor  
Peter Juhasz  
Hilal Lashuel  
Michael Ahlijanian, Henrik Zetterberg, Andreas Jeromin

**The Michael J. Fox Foundation for Parkinson’s Research**  
**PARKINSON’S PROGRESSION MARKERS INITIATIVE**

<table>
<thead>
<tr>
<th>Description of sub-species</th>
<th>Peptide Modification</th>
<th>Peptide Sequence</th>
<th>LOQ (fmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>aSyn_13-21.2</td>
<td>WT</td>
<td>EGVVAAEK</td>
<td>1</td>
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<tr>
<td>aSyn_33-43.2</td>
<td>WT, missed cleavage</td>
<td>TKEGVLVYGSK</td>
<td>2.5</td>
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<tr>
<td>aSyn_35-43_pY79</td>
<td>pY79</td>
<td>EGVpLVYGSK</td>
<td>0.5</td>
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<tr>
<td>aSyn_35-43_rY79</td>
<td>rY79</td>
<td>EGVLYVYGSK</td>
<td>0.1</td>
</tr>
<tr>
<td>aSyn_46-58</td>
<td>WT</td>
<td>EGVLVYGSK</td>
<td>0.1</td>
</tr>
<tr>
<td>aSyn_56-68</td>
<td>WT</td>
<td>TVEGAGSAATGFVK</td>
<td>10</td>
</tr>
<tr>
<td>aSyn_81-96_pS87</td>
<td>pS87</td>
<td>DNEYEMPSEEYQ</td>
<td>25</td>
</tr>
<tr>
<td>aSyn_81-96_pS87</td>
<td>pS87</td>
<td>DNEYEMPSEEYQ</td>
<td>25</td>
</tr>
<tr>
<td>aSyn112_131-140</td>
<td>Isoform 2.5</td>
<td>EGVQYEPEA</td>
<td>5</td>
</tr>
<tr>
<td>aSyn1126_35-40-55-58</td>
<td>Isoform 2.4</td>
<td>EGVLYVYAEK</td>
<td>0.1</td>
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<tr>
<td>aSyn119_103-119_m116</td>
<td>Trunc @ 119, MeO</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>10</td>
</tr>
<tr>
<td>aSyn119_103-119</td>
<td>Trunc @ 119</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>5</td>
</tr>
<tr>
<td>aSyn120_103-120_m116</td>
<td>Trunc @ 120, MeO</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>25</td>
</tr>
<tr>
<td>aSyn120_103-120</td>
<td>Trunc @ 120</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>25</td>
</tr>
<tr>
<td>aSyn122_103-122_m116</td>
<td>Trunc @ 122, MeO</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>10</td>
</tr>
<tr>
<td>aSyn122_103-122</td>
<td>Trunc @ 122</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>10</td>
</tr>
<tr>
<td>aSyn123_103-123_m116</td>
<td>Trunc @ 123, MeO</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>5</td>
</tr>
<tr>
<td>aSyn123_103-123</td>
<td>Trunc @ 123</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>5</td>
</tr>
<tr>
<td>aSyn124_103-124_m116</td>
<td>Trunc @ 124, MeO</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>5</td>
</tr>
<tr>
<td>aSyn124_103-124</td>
<td>Trunc @ 124</td>
<td>NEEGAPOQEGILEDMPVD</td>
<td>5</td>
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<tr>
<td>aSyn_46-58_A53T</td>
<td>Mutation A53T</td>
<td>EGVHGVTTVAEK</td>
<td>0.5</td>
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<tr>
<td>aSyn_46-58_A53T</td>
<td>Mutation A53T</td>
<td>EGVHGVTTVAEK</td>
<td>0.5</td>
</tr>
</tbody>
</table>

**PI:** Brit Mollenhauer  
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Natural History of Parkinson disease

Neuron Function

Pre-diagnostic

Symptomatic Diagnosis

Current Biomarker Studies

Clinical course heterogeneity

RBD

Hyposmia

PPMI

DeNoPa

Honolulu Asia Aging Study

SINBAR

PARS

EIPARK

TREND

RBD

Hyposmics

LRRK-2

SINBAR

PARS

TREND

DeNoPa substudy

P-PPMI

RBD

Hyposmics

LRRK-2

EPIPARK

PRIPS

DeNoPa

substudy

Honolulu Asia Aging Study

SINBAR

PARS

EIPARK

TREND

DeNoPa

substudy

P-PPMI
www.ppmi-info.org
Acknowledgement

Clinic (KS)
Claudia Trenkwalder
Friederike Sixel-Döring
Jens Ebentheuer
Monica Canelo

Lab
Niels Kruse (Gö)
Birgit Otte (Gö/KS)
Olivia Steuer (KS)

Sleep Lab
Friederike Sixel-Döring (KS)
Andrea Wegener (KS)
Norbert Drescher (KS)

MRI/Imaging
Niels Focke (Gö)

Neuropsychology
Martina Schaumburg (KS)

Statistical analysis
Johannes Zimmermann (KS)
Joe J. Locascio (Boston)

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Omar El-Agnaf, Al-Ain, United Arab Emirates
Karsten Hiller, Luxemburg
Christine Klein/Katja Lohmann, Univ. Lübeck
Ralf Kohnen†, RPS, Nürnberg
Hilal Lashuel, Adrian Schmid, Lausanne, Switzerland
Katrin Marcus, Medical Proteomcenter Bochum
Tiago Outeiro, Univ. Göttingen
Michael Schlossmacher, OHRI, Ottawa, Canada
Anja Schneider, Univ. Göttingen
Peggy Taylor, BioLegend, USA
Eugeen Vanmechelen und Hugo Vanstichelen, ADx Neurosciences, Ghent, Belgium
Heike Wersching, Klaus Berger, Münster University
Juliane Winkelmann, Center of Human Genetics, Munich/Stanford University
Henrik Zetterberg, Gothenborg University, Sweden

IITs from TEVA Pharma,
Boehringer-Ingelheim,
GE, DiaGenics