Longitudinal CSF biomarkers in patients with early Parkinson disease and healthy controls

ABSTRACT

Objective: To analyze longitudinal levels of CSF biomarkers in drug-naive patients with Parkinson disease (PD) and healthy controls (HC), examine the extent to which these biomarker changes relate to clinical measures of PD, and identify what may influence them.

Methods: CSF a-synuclein (a-syn), total and phosphorylated tau (t- and p-tau), and b-amyloid 1-42 (Ab42) were measured at baseline and 6 and 12 months in 173 patients with PD and 112 matched HC in the international multicenter Parkinson’s Progression Marker Initiative. Baseline clinical and demographic variables, PD medications, neuroimaging, and genetic variables were evaluated as potential predictors of CSF biomarker changes.

Results: CSF biomarkers were stable over 6 and 12 months, and there was a small but significant increase in CSF Ab42 in both patients with PD and HC from baseline to 12 months. The t-tau remained stable. The p-tau increased marginally more in patients with PD than in HC. a-syn remained relatively stable in patients with PD and HC. Ratios of p-tau/t-tau increased, while t-tau/Ab42 decreased over 12 months in patients with PD. CSF biomarker changes did not correlate with changes in Movement Disorder Society-sponsored revision of the Unified Parkinson’s Disease Rating Scale motor scores or dopamine imaging. CSF a-syn levels at 12 months were lower in patients with PD treated with dopamine replacement therapy, especially dopamine agonists.

Conclusions: These core CSF biomarkers remained stable over 6 and 12 months in patients with early PD and HC. PD medication use may influence CSF a-syn. Novel biomarkers are needed to better profile progressive neurodegeneration in PD. Neurology® 2017;89:1959-1969

GLOSSARY

a-syn = a-synuclein; Ab42 = b-amyloid 1-42; AD = Alzheimer disease; HC = healthy controls; LED = levodopa equivalent dose; MDS-UPDRS = Movement Disorder Society-sponsored revision of the Unified Parkinson’s Disease Rating Scale; MoCA = Montreal Cognitive Assessment; p-tau = phosphorylated tau protein; PD = Parkinson disease; Penn = University of Pennsylvania; PPMI = Parkinson’s Progression Biomarker Initiative; RBD = REM sleep behavior disorder; RBD-SQ = REM sleep behavior disorder screening questionnaire; t-tau = total tau protein.

Intracellular accumulation of a-synuclein (a-syn) aggregates, neuronal dysfunction and loss, and synaptic changes are the neuropathologic hallmarks of Parkinson disease (PD). Mutations and duplications in the a-syn encoding gene (SNCA) are associated with autosomal dominantly inherited PD, providing further support for a central role of a-syn in PD. Recent evidence suggests that transcellular spread of aggregated or misfolded a-syn may contribute to progression via the extracellular space. This raises the possibility that the quantification of a-syn in extracellular fluids may be a marker for PD diagnosis and progression. Total tau (t-tau) and phosphorylated tau (p-tau)
protein, as well as β-amyloid 1-42 (Aβ42), correlate with key pathologic features in Alzheimer disease (AD). These proteins have been shown to be relevant in PD neurodegeneration with an association between the microtubule-associated protein tau (MAPT) gene with PD and a known overlapping pathology (with AD). In cross-sectional studies, levels of α-syn in CSF are decreased in PD and related disorders. This decrease of CSF α-syn and changes in t-tau and p-tau protein and Aβ42 were recently replicated in the large multicenter Parkinson’s Progression Biomarker Initiative (PPMI). Longitudinal changes in levels of CSF α-syn and other biomarkers in PD were examined in other cohorts with suggestions that CSF α-syn may increase over time or in those with more severe PD. Understanding the dynamics of changes in biomarkers may advance our understanding of the pathobiology of the disease course, identify contributions of different pathologies to progression, and can provide benchmark data for the design and interpretation of disease-modifying clinical trials that use biomarkers for participant enrollment or as outcome measures.

We therefore analyzed the levels of α-syn, tau, p-tau, and Aβ42 in CSF samples of patients with PD and healthy controls (HC) at baseline and 6- and 12-month follow-up in the PPMI cohort. We hypothesized that these core CSF biomarkers would be stable in patients with PD and HC and would correlate with clinical or ¹²³I-ioflupane dopamine transporter imaging (DaTscan) indices of disease progression.

METHODS Participants. People with recently diagnosed untreated PD were enrolled in PPMI. PPMI is an ongoing prospective longitudinal, observational, international multicenter study that aims to identify biomarkers for the progression of PD. As described previously, newly diagnosed, drug-naive patients with PD (n = 423), age- and sex-matched HC (n = 196), and participants with scans without evidence of dopaminergic deficit syndrome (n = 60) were included in the study. Recruitment took place between June 2010 and May 2013, in 21 PD centers in the United States and Europe in accordance with PPMI protocols. The criteria for enrollment between June 2010 and May 2013 for participants with PD were (1) age over 30 years; (2) presence of 2 of the following: bradykinesia, rigidity, and resting tremor, or presence of an asymmetric resting tremor, or asymmetric bradykinesia; (3) diagnosis recently made within the last 24 months; (4) PD drug naivety; and (5) dopamine transporter deficit in the putamen on diagnosis recently made within the last 24 months; (4) PD drug of an asymmetric resting tremor, or asymmetric bradykinesia; (3) People with recently diagnosed untreated PD were free of dopamine-related medications. Use of medications for PD was recorded at the 6- and 12-month visits, and is expressed as levodopa equivalent doses (LEDs).

Clinical assessment measures. The clinical assessment battery is described on the PPMI website. In brief, motor assessment was performed with the Movement Disorder Society–sponsored revision of the Unified Parkinson’s Disease Rating Scale (MDS-UPDRS) III and total score. At baseline, all participants with PD were free of dopamine-related medications. Use of medications for PD was recorded at the 6- and 12-month visits, and is expressed as levodopa equivalent doses (LEDs).

Cognitive testing comprised screening with the Montreal Cognitive Assessment (MoCA) and the Hopkins Verbal Learning Test–revised, processing speed/attention was assessed using the Symbol Digit Modality Test, executive function/working memory was assessed with the Wechsler Memory Scale III Letter-Number Sequencing Test, and visuospatial abilities were assessed with the Benton Judgment of Line Orientation test. The REM sleep behavior disorder (RBD) screening questionnaire (RBD-SQ) was used to assess RBD.

Dopamine SPECT imaging. Dopamine imaging was performed by DaTscan using standardized methods, as described. We analyzed whether quantitative DaTscan measures of caudate, putamen, or striatal uptake were related to CSF biomarker changes.

Genetic variables. To examine whether selected genetic variants were associated with CSF biomarkers, we used data for APOE genotypes and single nucleotide polymorphisms related to SNCA. These were measured by the PPMI Genetics Core as previously described.

Statistical analysis. All analyses are based on data retrieved from the PPMI website, when all biomarkers for the 6- and 12-month follow-up periods were available on January 19, 2016. All statistical analyses were performed using SAS 9.4 (SAS Institute Inc., Cary, NC). All tests performed using the CSF biomarkers were rank-based. t Tests or χ² tests were used to compare baseline demographic and clinical variables in participants with longitudinal CSF data vs participants who only had baseline CSF data; these comparisons were performed separately in patients with PD and controls. Nonparametric tests were used where specified in the tables. Repeated-measures linear mixed models were used to test for changes in CSF biomarker levels from...
### Table 1 Baseline clinical characteristics of patients with Parkinson disease (PD) and healthy controls (HC) with longitudinal CSF data

| Characteristics                                      | Patients with PD (n = 173) | HC (n = 112) |
|------------------------------------------------------|----------------------------|--------------|
| Age at baseline lumbar puncture, y                   | n                          | 173          |
|                                                      | Mean (SD)                  | 60.91 (9.3)  |
|                                                      | 95% CI                     | 59.5-62.3    |
| Sex, n (%)                                           | n                          | 169          |
|                                                      | Mean (SD)                  | 58.79 (9.7)  |
|                                                      | 95% CI                     | 57.3-60.3    |
| MDS-UPDRS part III (motor score)                     | n                          | 173          |
|                                                      | Mean (SD)                  | 21.86 (8.6)  |
|                                                      | 95% CI                     | 20.6-23.2    |
| MDS-UPDRS total score                                | n                          | 173          |
|                                                      | Mean (SD)                  | 33.61 (13.4) |
|                                                      | 95% CI                     | 31.6-35.6    |
| TD/non-TD classification, n (%)                       |                              |              |
| TD                                                   | n                          | 126 (73)     |
|                                                      | Mean (SD)                  | 8.29 (2.4)   |
|                                                      | 95% CI                     | 7.9-8.7      |
| Non-TD                                               | n                          | 47 (27)      |
|                                                      | Mean (SD)                  | 9.41 (2.3)   |
|                                                      | 95% CI                     | 9.0-9.8      |
| HVLT total recall                                    | n                          | 173          |
|                                                      | Mean (SD)                  | 24.43 (4.5)  |
|                                                      | 95% CI                     | 23.8-25.1    |
| HVLT delayed recall                                  | n                          | 173          |
|                                                      | Mean (SD)                  | 8.90 (2.4)   |
|                                                      | 95% CI                     | 8.0-8.7      |
| HVLT discrimination recognition                      | n                          | 173          |
|                                                      | Mean (SD)                  | 9.62 (2.5)   |
|                                                      | 95% CI                     | 9.3-10.0     |
| MoCA                                                 | n                          | 173          |
|                                                      | Mean (SD)                  | 27.04 (2.2)  |
|                                                      | 95% CI                     | 26.7-27.4    |
| SDMT                                                 | n                          | 173          |
|                                                      | Mean (SD)                  | 41.43 (8.8)  |
|                                                      | 95% CI                     | 40.1-42.8    |
| LNS                                                  | n                          | 173          |
|                                                      | Mean (SD)                  | 124 (72)     |
|                                                      | 95% CI                     | 120-128      |
|                                                      | Mean (SD)                  | 70 (63)      |
|                                                      | 95% CI                     | 66-74        |
|                                                      | Women                      | 49 (28)      |
|                                                      | 95% CI                     | 45-53        |
|                                                      | Mean (SD)                  | 169 NA       |
|                                                      | 95% CI                     | 168 NA       |
|                                                      | Mean (SD)                  | 57.2 NA      |
|                                                      | 95% CI                     | 56.9 NA      |
|                                                      | Sex (men/female)            | 59.5 NA      |
|                                                      | 95% CI                     | 59.2 NA      |
|                                                      | Age at PD onset, y          | 40.1 NA      |
|                                                      | 95% CI                     | 39.8 NA      |
|                                                      | Age at baseline lumbar puncture, y | 173 112 |
|                                                      | 95% CI                     | 169 NA       |
|                                                      | Mean (SD)                  | 33.61 (13.4) |
|                                                      | 95% CI                     | 31.6-35.6    |
|                                                      | Sex (men/female)            | 57.3 NA      |
|                                                      | 95% CI                     | 57.0 NA      |
|                                                      | Age at PD onset, y          | 40.1 NA      |
|                                                      | 95% CI                     | 39.8 NA      |
|                                                      | Age at baseline lumbar puncture, y | 173 112 |
|                                                      | 95% CI                     | 169 NA       |
|                                                      | Mean (SD)                  | 33.61 (13.4) |
|                                                      | 95% CI                     | 31.6-35.6    |
|                                                      | Sex (men/female)            | 57.3 NA      |
|                                                      | 95% CI                     | 57.0 NA      |
|                                                      | Age at PD onset, y          | 40.1 NA      |
|                                                      | 95% CI                     | 39.8 NA      |
|                                                      | Age at baseline lumbar puncture, y | 173 112 |
|                                                      | 95% CI                     | 169 NA       |
|                                                      | Mean (SD)                  | 33.61 (13.4) |
|                                                      | 95% CI                     | 31.6-35.6    |
|                                                      | Sex (men/female)            | 57.3 NA      |
|                                                      | 95% CI                     | 57.0 NA      |

Continued
Table 1 Continued

|                | Patients with PD (n = 173) | HC (n = 112) |
|----------------|---------------------------|--------------|
| **Mean (SD)**  | 10.84 (2.3)               | 10.86 (2.6)  |
| **95% CI**     | 10.5-11.2                 | 10.4-11.3    |
| **BJLO**       |                           |              |
| n              | 173                       | 112          |
| **Mean (SD)**  | 12.94 (2.1)               | 13.21 (1.9)  |
| **95% CI**     | 12.6-13.3                 | 12.9-13.6    |

Abbreviations: BJLO = Benton Judgment of Line Orientation test; CI = confidence interval; HVLT = Hopkins Verbal Learning Test; LNS = Letter-Number Sequencing Test; MDS-UPDRS = Movement Disorder Society-sponsored revision of the Unified Parkinson’s Disease Rating Scale; MoCA = Montreal Cognitive Assessment; NA = not applicable; SDMT = Symbol Digit Modality Test; TD = tremor dominant.

There were moderate and significant correlations among the 4 CSF markers, which are summarized in table e-6.

**DISCUSSION**

This multicenter longitudinal study evaluated core CSF biomarkers—including α-syn, Aβ42, t-tau, and p-tau levels—measured over 6 and 12 months in patients with de novo PD and healthy controls. The strengths of the data include quality control and standardization of recruitment of participants, support of clinical diagnosis through DaTscan imaging, rigorous clinical assessment, CSF and biosample collection, handling and central analysis according to established standardized operational procedures, and functional as well as structural brain image analysis, together with high recruitment numbers, retention rates, and rates of performance of longitudinal lumbar punctures. The diversity of enrollment sites is representative of a typical multicenter interventional study. Therefore, these data can serve as a benchmark for future intervention trials.

Overall, we show stability of all 4 biomarkers during 12 months of follow-up in de novo PD. Therefore, these CSF biomarkers do not mirror disease progression, in particular progressive striatoniatal degeneration as evaluated by clinical motor ratings (MDS-UPDRS III) and DaTscan measures. Whether these CSF biomarkers change over a longer time course, during more advanced stages of PD, or in relation to, for example, blood–brain barrier changes, can be reassessed once further PPMI biomarker analyses are conducted.

CSF α-syn assays measure the total physiologic protein rather than its select pathologic forms, and cellular events that lead to its release into extracellular CSF are not well-understood. The development of assays that measure other forms of α-syn such as Pser129 α-syn25 or α-syn oligomers23-24 may provide stronger indices of disease activity. Although it is possible that decreased levels of CSF α-syn in PD may normalize (increase) with effective neuroprotective therapy as target engagement, this will need to be tested in the setting of an effective intervention. CSF levels of t-tau and p-tau181 levels have been extensively studied in AD, where they are related to neuronal damage and neurofibrillary changes. They increase in the presymptomatic mild cognitive impairment stage, and remain stably elevated or even decrease slightly once the symptomatic phase with memory loss is present.25,26 It may therefore be important to similarly analyze people at risk for PD, such as asymptomatic mutation carriers and people with idiopathic nonmotor symptoms, such as RBD or hyposmia.27

Analyses of t-tau and p-tau proteins and Aβ42 in longitudinal CSF samples in 403 drug-naive patients...
Table 2  Movement Disorder Society–sponsored revision of the Unified Parkinson’s Disease Rating Scale (MDS-UPDRS), DaTscan levels, and CSF analytes ([α]-amyloid 1-42 [Aβ42], total tau protein [t-tau], phosphorylated tau protein [p-tau], α-synuclein [α-syn], and their ratios) at baseline and 6 and 12 months in patients with Parkinson disease (PD) and healthy controls (HC)

| Variable                  | Patients with PD                                                                 | HC                                                                 |
|---------------------------|----------------------------------------------------------------------------------|----------------------------------------------------------------------|
|                           | Baseline | Change at 6 months | Change at 12 months | p Value (baseline vs 6 months) | Baseline | Change at 6 months | Change at 12 months | p Value (baseline vs 6 months) | p Value (baseline vs 12 months) |
| **MDS-UPDRS III**         |          |                    |                    |                                 |          |                    |                    |                                 |                                |
| n                         | 173      | 173                | 148                | -0.0001                         |          |                    |                    |                                 |                                |
| Mean (SD)                 | 21.86 (8.6) | 3.41 (6.0)        | 4.46 (7.9)         | 1.38 (2.4)                      |          |                    |                    | 0.53 (2.0)                      |                                |
| Min, max                  | 20.6, 23.2 | 2.5, 4.3           | 3.2, 5.7           | 0.9, 1.8                        |          |                    |                    | 0.1, 0.9                        |                                |
| **Mean caudate**          |          |                    |                    | -0.0001                         |          |                    |                    |                                 |                                |
| n                         | 172      | NA                 | 169                |                                  |          |                    |                    |                                 |                                |
| Mean (SD)                 | 1.99 (0.549) | -0.22 (0.27)      | 2.98 (0.63)        |                                  |          |                    |                    |                                 |                                |
| Min, max                  | 1.90, 2.07 | -0.26, -0.18       | 2.86, 3.09         |                                  |          |                    |                    |                                 |                                |
| **Mean putamen**          |          |                    |                    | -0.0001                         |          |                    |                    |                                 |                                |
| n                         | 172      | NA                 | 169                |                                  |          |                    |                    |                                 |                                |
| Mean (SD)                 | 0.79 (0.28) | -0.11 (0.15)      | 2.09 (0.55)        |                                  |          |                    |                    |                                 |                                |
| Min, max                  | 0.75, 0.84 | -0.14, -0.10       | 1.99, 2.20         |                                  |          |                    |                    |                                 |                                |
| **Mean striatum**         |          |                    |                    | -0.0001                         |          |                    |                    |                                 |                                |
| n                         | 172      | NA                 | 169                |                                  |          |                    |                    |                                 |                                |
| Mean (SD)                 | 1.39 (0.39) | -0.17 (0.19)      | 2.53 (0.57)        |                                  |          |                    |                    |                                 |                                |
| Min, max                  | 1.33, 1.45 | -0.20, -0.14       | 2.43, 2.64         |                                  |          |                    |                    |                                 |                                |
| **Aβ42**                  |          | 0.33               | 0.1               | 0.09                             |          |                    |                    | 0.02                            |                                |
| n                         | 173      | 173                | 173                | 112                              |          |                    |                    | 112                             |                                |
| Median                    | 361.10   | 7.50               | 11.50              | 378.15                          |          |                    |                    | 6.15                            | 21.35                           |
| Min, max                  | 139.9, 670.0 | -242.2, 205.0    | -207.1, 316.8      | 88.8, 680.3                     | -230.3, 152.4        | -265.3, 190.0       |                                   | 112                             |                                |
| **t-tau**                 |          | 0.1                | 0.56               | 0.81                             |          |                    |                    | 0.30                            |                                |
| n                         | 171      | 171                | 170                | 110                              |          |                    |                    | 110                             |                                |
| Median                    | 38.70    | -1.20              | -0.15              | 44.65                           |          |                    |                    | -0.25                           | 0.70                            |
| Min, max                  | 15.6, 121.0 | -15.4, 21.8      | -28.4, 36.7        | 18.4, 188.2                     | -26.6, 35.4          | -27.8, 39.6         |                                   | 110                             |                                |
| **p-tau**                 |          | 0.32               | 0.001              | 0.19                             |          |                    |                    | 0.15                            |                                |
| n                         | 173      | 173                | 172                | 112                              |          |                    |                    | 112                             |                                |
| Median                    | 11.40    | 0.20               | 1.95               | 14.00                           |          |                    |                    | -0.35                           | 0.55                            |
| Min, max                  | 4.7, 39.7 | -21.7, 40.3        | -26.5, 46.6        | 6.1, 58.5                       | -24.3, 39.3          | -30.4, 74.6         |                                   | 112                             |                                |

Continued
| Variable | Patients with PD | | | | HC | | | | | p Value (baseline vs 6 months) | p Value (baseline vs 12 months) | p Value (baseline vs 6 months) | p Value (baseline vs 12 months) |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| | Baseline | Change at 6 months | Change at 12 months | | Baseline | Change at 6 months | Change at 12 months | | | | | |
| α-syn | 0.43 | 0.79 | 0.24 | 0.96 | 0.77 | 0.55 | | | | | |
| n | 173 | 172 | 173 | | 112 | 112 | 112 | | | | |
| Median | 1714.39 | 31.95 | 14.15 | | 1950.13 | 8.81 | −5.00 | | | | |
| Min, max | 332.93, 6694.55 | −4503.15, 1301.97 | −4602.02, 1540.90 | | 592.56, 5237.68 | −1912.52, 2258.43 | −2292.36, 1748.10 | | | | |
| p-tau/t-tau | 0.34 | 0.002 | 0.39 | 0.43 | | | | | | | |
| n | 82 | 81 | 82 | | 43 | 43 | 43 | | | | |
| Median | 1712.71 | 33.74 | 55.01 | | 1941.14 | 5.26 | −101.64 | | | | |
| Min, max | 581.17, 5110.77 | −2239.79, 837.42 | −2107.86, 1284.25 | | 683.94, 5237.68 | −1912.52, 1164.75 | −2292.36, 1529.98 | | | | |
| t-tau/A | 0.007 | 0.003 | 0.80 | 0.57 | | | | | | | |
| n | 171 | 171 | 170 | | 110 | 110 | 110 | | | | |
| Median | 0.11 | −0.01 | −0.01 | | 0.11 | −0.002 | −0.003 | | | | |
| Min, max | 0.06, 0.53 | −0.06, 0.24 | −0.15, 0.27 | | 0.071, 2.12 | −0.278, 0.08 | −0.199, 0.15 | | | | |
| p-tau/A42 | 0.42 | 0.01 | 0.04 | 0.73 | | | | | | | |
| n | 173 | 173 | 172 | | 112 | 112 | 112 | | | | |
| Median | 0.03 | 0.00 | 0.01 | | 0.04 | −0.00 | 0.00 | | | | |
| Min, max | 0.01, 0.17 | −0.07, 0.09 | −0.08, 0.23 | | 0.02, 0.66 | −0.23, 0.07 | −0.19, 0.39 | | | | |
| t-tau/α-syn | 0.09 | 0.61 | 0.32 | 0.47 | | | | | | | |
| n | 171 | 170 | 170 | | 110 | 110 | 110 | | | | |
| Median | 0.02 | −0.001 | −0.001 | | 0.02 | −0.001 | 0.00 | | | | |
| Min, max | 0.01, 0.07 | −0.05, 0.02 | −0.04, 0.02 | | 0.01, 0.06 | −0.03, 0.02 | −0.03, 0.02 | | | | |
| p-tau/α-syn* | 0.12 | 0.21 | 0.38 | 0.70 | | | | | | | |
| n | 82 | 81 | 81 | | 43 | 43 | 43 | | | | |
| Median | 0.02 | −0.00 | −0.00 | | 0.024 | −0.001 | 0.000 | | | | |
| Min, max | 0.01, 0.05 | −0.03, 0.02 | −0.02, 0.02 | | 0.01, 0.06 | −0.03, 0.01 | −0.03, 0.02 | | | | |

Abbreviation: NA = not applicable.

p Values are based on the ranks of the biological variables. DaTscan is not performed at 6 months in patients with PD, and is performed only at baseline in HC.

*Subset of participants with hemoglobin >200 ng/mL at all time points, excluding those with missing hemoglobin values at one or more time points.
| Variables | Patients with PD | HC | p Value (PD vs HC) |
|-----------|-----------------|----|-------------------|
| Aβ42      |                 |    | 0.134             |
| n         | 173             | 173| 173              |
| Median    | 361.10          | 361.70| 375.50|
| Min, max  | 139.90, 670.00  | 129.30, 687.00| 144.10, 732.50|
| t-tau     |                 |    | 0.0004            |
| n         | 171             | 173| 172              |
| Median    | 38.70           | 37.40| 39.05|
| Min, max  | 15.60, 121.00   | 15.60, 134.70| 16.60, 128.80|
| p-tau     |                 |    | 0.006             |
| n         | 173             | 173| 172              |
| Median    | 11.40           | 11.50| 14.10|
| Min, max  | 4.70, 39.70     | 510, 56.30| 5.40, 61.80|
| α-syn     |                 |    | 0.002             |
| n         | 173             | 172| 173              |
| Median    | 1714.39         | 1779.73| 1720.77|
| Min, max  | 332.93, 6694.55 | 472.92, 4659.05| 352.36, 5157.08|
| α-syn*    |                 |    | 0.109             |
| n         | 82              | 81 | 82               |
| Median    | 1712.71         | 1802.91| 1816.15|
| Min, max  | 581.17, 5110.77 | 707.06, 4264.73| 797.87, 5157.08|
| p-tau/t-tau|                |    | 0.215             |
| n         | 171             | 173| 171              |
| Median    | 0.30            | 0.29| 0.340            |
| Min, max  | 0.05, 0.88      | 0.07, 2.48| 0.07, 0.79|
| t-tau/Aβ42|                 |    | 0.002             |
| n         | 171             | 173| 172              |
| Median    | 0.11            | 0.10| 0.10            |
| Min, max  | 0.06, 0.53      | 0.06, 0.51| 0.071, 2.12|
| p-tau/α-syn|                |    | 0.085             |
| n         | 173             | 173| 172              |
| Median    | 0.03            | 0.03| 0.04            |
| Min, max  | 0.01, 0.17      | 0.01, 0.28| 0.02, 0.66|
| t-tau/α-syn*|               |    | 0.202             |
| n         | 171             | 172| 172              |
| Median    | 0.02            | 0.02| 0.02            |
| Min, max  | 0.01, 0.07      | 0.01, 0.06| 0.01, 0.06|
| t-tau/α-syn*|               |    | 0.515             |
| n         | 82              | 81 | 81               |
| Median    | 0.02            | 0.02| 0.02            |
| Min, max  | 0.01, 0.05      | 0.01, 0.06| 0.01, 0.06|

*p Values are based on the ranks of the biologic variables.

*Subset of participants with hemoglobin <200 ng/mL at all time points; excludes those missing hemoglobin values at one or more time points.
with PD at enrollment in the Deprenyl and Tocopherol Antioxidative Therapy of PD (DATATOP) placebo-controlled clinical trial revealed a slight but significant positive correlation between the rate of change in t-tau or t-tau/Aβ levels and changes in Unified Parkinson’s Disease Rating Scale scores. In the PPMI cohort, the correlation between clinical progression by total MDS-UPDRS scores and changes of CSF α-syn after 6 and 12 months of observation supports a pathophysiologic connection of CSF α-syn levels with motor progression, albeit weak. However, this is not directly related to measures of presynaptic dopamine integrity in the basal ganglia by DaTscan.

Since the PD phenotype is very heterogeneous, different subtypes could show different biomarker

### Table 4: Correlation between change in CSF biomarkers and change in the Movement Disorder Society-sponsored revision of the Unified Parkinson’s Disease Rating Scale (MDS-UPDRS) part III motor score and the total score in patients with Parkinson disease (PD) and healthy controls (HC)

|                      | Patients with PD | HC |
|----------------------|-----------------|----|
|                      | Change at 6 months | Change at 12 months |     | Change at 12 months |
|                      | Spearman correlation coefficient | p Value | Spearman correlation coefficient | p Value | Spearman correlation coefficient | p Value |
| Correlation with Aβ42|                 |                |                         |                         |                         |
| MDS-UPDRS part III score | −0.08 | 0.296 | 0.01 | 0.879 | −0.11 | 0.266 |
| MDS-UPDRS total score   | −0.13 | 0.082 | −0.01 | 0.895 | −0.01 | 0.916 |
| Correlation with t-tau |                 |                |                         |                         |                         |
| MDS-UPDRS part III score | −0.02 | 0.772 | 0.03 | 0.713 | 0.09 | 0.369 |
| MDS-UPDRS total score   | −0.07 | 0.369 | −0.03 | 0.718 | 0.09 | 0.347 |
| Correlation with p-tau |                 |                |                         |                         |                         |
| MDS-UPDRS part III score | 0.11 | 0.162 | 0.05 | 0.563 | −0.00 | 0.999 |
| MDS-UPDRS total score   | 0.09 | 0.247 | 0.06 | 0.471 | −0.01 | 0.898 |
| Correlation with α-syn |                 |                |                         |                         |                         |
| MDS-UPDRS part III score | 0.12 | 0.117 | 0.12 | 0.118 | 0.13 | 0.168 |
| MDS-UPDRS total score   | 0.15 | 0.047 | 0.17 | 0.029 | 0.07 | 0.470 |
| Correlation with α-syn (low Hgb) |               |                |                         |                         |                         |
| MDS-UPDRS part III score | 0.17 | 0.120 | −0.02 | 0.882 | 0.10 | 0.515 |
| MDS-UPDRS total score   | 0.19 | 0.097 | −0.03 | 0.808 | −0.12 | 0.460 |
| Correlation with p-tau/τ-tau |               |                |                         |                         |                         |
| MDS-UPDRS part III score | 0.09 | 0.252 | 0.01 | 0.944 | −0.04 | 0.650 |
| MDS-UPDRS total score   | 0.08 | 0.304 | 0.04 | 0.587 | −0.06 | 0.513 |
| Correlation with t-tau/α42 |               |                |                         |                         |                         |
| MDS-UPDRS part III score | 0.08 | 0.304 | −0.03 | 0.737 | 0.19 | 0.047 |
| MDS-UPDRS total score   | 0.04 | 0.611 | −0.06 | 0.466 | 0.15 | 0.110 |
| Correlation with p-tau/α42 |               |                |                         |                         |                         |
| MDS-UPDRS part III score | 0.13 | 0.090 | 0.034 | 0.667 | 0.03 | 0.782 |
| MDS-UPDRS total score   | 0.1291 | 0.090 | 0.056 | 0.475 | −0.03 | 0.813 |
| Correlation with t-tau/α-syn |               |                |                         |                         |                         |
| MDS-UPDRS part III score | −0.06 | 0.446 | −0.06 | 0.475 | −0.09 | 0.369 |
| MDS-UPDRS total score   | −0.11 | 0.147 | −0.15 | 0.055 | −0.07 | 0.480 |
| Correlation with t-tau/α-syn (low Hgb) |               |                |                         |                         |                         |
| MDS-UPDRS part III score | −0.034 | 0.756 | 0.22 | 0.050 | −0.04 | 0.787 |
| MDS-UPDRS total score   | −0.05 | 0.643 | 0.22 | 0.051 | 0.01 | 0.937 |

**Abbreviations:** α-syn = α-synuclein; Aβ42 = β-amyloid 1-42; p-tau = phosphorylated tau protein; Hgb = hemoglobin; t-tau = total tau protein.

Controls did not complete MDS-UPDRS at 6 months.

*Subset of participants with Hgb <200 ng/mL at all time points; excludes those missing Hgb values at one or more time points.*
Table 5 Longitudinal relationship between CSF biomarkers and Parkinson disease (PD) medications in patients with PD (calculated as levodopa equivalent dosages [LED] as published)

| Variable                                                      | Patients with PD | p Value |
|---------------------------------------------------------------|------------------|---------|
| Relationship with \(\text{A\beta} 42\)                        |                  |         |
| PD medication use                                             | \(-4.07 (-29.38\) to 21.24) | 0.750   |
| Total LED                                                     | 0.08 (-0.01 to 0.16) | 0.074   |
| LED subtotal—dopamine replacement                            | 0.08 (-0.02 to 0.18) | 0.114   |
| LED subtotal—dopamine agonants                               | -0.00 (-0.18 to 0.18) | 0.969   |
| Relationship with \(\tau\)-tau                               |                  |         |
| PD medication use                                             | 5.76 (-8.81 to 20.33) | 0.434   |
| Total LED                                                     | 0.03 (-0.01 to 0.08) | 0.164   |
| LED subtotal—dopamine replacement                            | 0.03 (-0.03 to 0.09) | 0.315   |
| LED subtotal—dopamine agonants                               | 0.04 (-0.06 to 0.14) | 0.400   |
| Relationship with \(\alpha\)-syn                              |                  |         |
| PD medication use                                             | -12.31 (-49.38 to 24.76) | 0.510   |
| Total LED                                                     | 0.06 (-0.07 to 0.18) | 0.365   |
| LED subtotal—dopamine replacement                            | 0.02 (-0.13 to 0.17) | 0.808   |
| LED subtotal—dopamine agonants                               | 0.13 (-0.14 to 0.39) | 0.340   |
| Relationship with \(\tau\)-tau                               |                  |         |
| PD medication use                                             | -28.54 (-48.40 to -8.69) | 0.005   |
| Total LED                                                     | -0.06 (-0.12 to 0.01) | 0.073   |
| LED subtotal—dopamine replacement                            | 0.02 (-0.06 to 0.10) | 0.560   |
| LED subtotal—dopamine agonants                               | -0.28 (-0.41 to -0.14) | 0.0001  |
| Relationship with \(\alpha\)-syn                            |                  |         |
| PD medication use                                             | -43.24 (-71.34 to -15.15) | 0.004   |
| Total LED                                                     | -0.09 (-0.19 to 0.01) | 0.077   |
| LED subtotal—dopamine replacement                            | 0.07 (-0.05 to 0.19) | 0.260   |
| LED subtotal—dopamine agonants                               | -0.42 (-0.59 to -0.25) | 0.0001  |
| Relationship with \(\tau\)-tau/\(\tau\)-tau                 |                  |         |
| PD medication use                                             | -7.10 (-45.86 to 31.65) | 0.716   |
| Total LED                                                     | 0.05 (-0.08 to 0.18) | 0.407   |
| LED subtotal—dopamine replacement                            | -0.01 (-0.17 to 0.14) | 0.878   |
| LED subtotal—dopamine agonants                               | 0.13 (-0.15 to 0.40) | 0.373   |
| Relationship with \(\tau\)-tau/\(\text{A\beta} 42\)          |                  |         |
| PD medication use                                             | 9.59 (-17.64 to 36.81) | 0.485   |
| Total LED                                                     | -0.01 (-0.10 to 0.08) | 0.803   |
| LED subtotal—dopamine replacement                            | -0.01 (-0.12 to 0.10) | 0.862   |
| LED subtotal—dopamine agonants                               | 0.04 (-0.15 to 0.23) | 0.682   |
| Relationship with \(\tau\)-tau/\(\text{A\beta} 42\)          |                  |         |
| PD medication use                                             | -11.09 (-47.81 to 25.62) | 0.549   |
| Total LED                                                     | 0.04 (-0.09 to 0.16) | 0.565   |
| LED subtotal—dopamine replacement                            | -0.01 (-0.16 to 0.14) | 0.891   |
| LED subtotal—dopamine agonants                               | 0.17 (-0.09 to 0.42) | 0.212   |
| Relationship with t-tau/\(\alpha\)-syn                        |                  |         |
| PD medication use                                             | 39.36 (8.42 to 70.29) | 0.013   |

Continued...
Values are based on the ranks of the biologic variables.

| Variable                                                                 | Estimate (95% CI) | p Value |
|--------------------------------------------------------------------------|-------------------|---------|
| Total LED                                                                | 0.10 (-0.00 to 0.20) | 0.061   |
| LED subtotal—dopamine replacement                                        | -0.00 (-0.13 to 0.12) | 0.991   |
| LED subtotal—dopamine agonists                                           | 0.36 (0.14 to 0.58) | 0.001   |
| Relationship with t-tau/α-synβ                                            | 0.27 (0.11 to 0.43) | 0.400   |
| PD medication use                                                         | 68.10 (24.76 to 111.44) | 0.003   |
| Total LED                                                                | 0.16 (0.01 to 0.32) | 0.044   |
| LED subtotal—dopamine replacement                                        | -0.08 (-0.27 to 0.11) | 0.400   |
| LED subtotal—dopamine agonists                                           | 0.67 (0.41 to 0.94) | <0.0001 |

Abbreviations: α-syn = α-synuclein; α42 = α-amyloid; CI = confidence interval; p-tau = phosphorylated tau; t-tau = total tau.

p Values are based on the ranks of the biologic variables.

*Subset of participants with hemoglobin <200 ng/mL at all time points; excludes those missing hemoglobin values at one or more time points.

AUTHOR CONTRIBUTIONS
B.M., D.G., and C.S.C. designed the study and were responsible for data processing. C.J.C.-G. and C.S.C. oversaw all statistical analyses. P.T., L.M.S., J.Q.T., and A.S. were involved in sample analyses and data interpretation. M.F. and K.M. oversaw patient recruitment and assisted in the interpretation of data. B.M., C.J.C.-G., and D.G. wrote the manuscript. C.S.C., P.T., L.M.S., J.Q.T., A.S., M.F., and K.M. coordinated the manuscript. B.M., D.G., and K.M. had full access to the clinical primary data. All authors had access to the data generated in the study including the statistical analysis and decided to submit the paper for publication.

ACKNOWLEDGMENT
The authors thank the Michael J. Fox Foundation, their PPMI colleagues, and the individuals who participated in this study.

STUDY FUNDING
PPMI is sponsored by the Michael J. Fox Foundation for Parkinson’s Research (MJFF) and is cofunded by MJFF, AbbVie, Avid Radiopharmaceuticals, Biogen Idec, Bristol-Myers Squibb, Covance, Eli Lilly & Co., F. Hoffman-La Roche, Ltd., GE Healthcare, Genentech, GlaxoSmithKline, Lundbeck, Merck, MesaScale, Piramal, Pfizer, and UCBB. This study is funded by The MJFF and funding partners, including Abbott, Biogen Idec, F. Hoffman-La Roche Ltd., GE Healthcare, Genentech, and Pfizer Inc. J.Q.T. is supported in part by P50 NS053488.

DISCLOSURE
B. Mollenhauer has received independent research grants from TEVA-Pharma, Desitin, Boehringer Ingelheim, and GE Healthcare; honoraria for consultancy from Bayer Schering Pharma AG, Roche, AbbVie, TEVA-Pharma, and Biogen; and for presentations from GlaxoSmithKline, Orion Pharma, and TEVA-Pharma; and travel costs from TEVA-Pharma. B.M. is a member of the executive steering committee of the Parkinson’s Progression Marking Initiative and the Systemic Synuclein Sampling Study of the Michael J. Fox Foundation for Parkinson’s Research (MJFF); has received grants from the BMBF, EU, Parkinson Fonds Deutschland, Deutsche Parkinson Vereinigung, MJFF, and Stiftverband für die deutsche Wissenschaft; has had scientific collaborations with Roche, Bristol Myers Squibb, Eli Lilly, Covance, and Biogen. C. Caspell-Garcia served as a consultant receiving fees from MJFF; and received research funding from The Michael J. Fox Foundation for Parkinson’s Research. C. Coffey served as a consultant receiving fees from MJFF; and received research funding from NINDS, NHLBI, and MJFF.

P. Taylor is an employee of Bio.legend. L. Shaw receives funding from the NIH/NIA, U19 AG049494, from MJFF, Eli Lilly, and Roche; honours for consultancy from Novartis and Eli Lilly; and travel expenses from Eli Lilly and Roche and honorarium from Eli Lilly and Novartis; and has provided QC oversight for immunohaspsy produced by Fujirebio and Roche as part of the ADNI and PPMI studies. J. Trojanowski may accrue revenue in the future on patents submitted by the University of Pennsylvania wherein he is coinventor and received revenue from the sale of Avid to Eli Lilly as coinventor on imaging-related patents submitted by the University of Pennsylvania; and receives research support from the NIH, GSK, Janssen, Biogen, and several nonprofits. A. Singleton was supported by grant 2014AG00949-06 from the Intramural Research Program, National Institute on Aging, NIH. J. H. H. was supported by grant MRC 2014009392 from the National Research Foundation of Korea, Ministry of Science, ICT, and Future Planning. M. Frasier is an employee of MJFF. K. Marek is a consultant for Pfizer, GE Healthcare, Merck, Lilly, BMS, Piramal, Prothena, Neuropharma, Eli Lilly, and Roche; receives funding for the following grants W81XWH-06-1-0678, Establishing an ‘at risk’ cohort for Parkinson Disease Neutroprevention using olfactory testing and DAT imaging, DOD, Investigator 10/06/09/30/15; Parkinson’s Progression Marker Initiative, Michael J. Fox Foundation, Principal Investigator 6/15/09-6/14/18; DAT imaging in LRRK2 family members, the Michael J. Fox Foundation, Principal Investigator 1/15/10-3/14/15; and has ownership in Molecular NeuroImaging, LLC. D. Galasko is supported by NIH grant AG05131 and by the Michael J. Fox Foundation; and has provided consultation for TV Pharmaceuticals, Eli Lilly, Inc., and Proclara, Inc. Go to Neurology.org for full disclosures.

Received February 12, 2017. Accepted in final form August 24, 2017.

REFERENCES
1. Klein C, Westenberger A. Genetics of Parkinson’s disease. Cold Spring Harb Perspect Med 2012;2:a008888.
2. Breiteneder J, Del Tredici K, Lee VM, Trojanowski JQ. Spreading of pathology in neurodegenerative diseases: a focus on human studies. Nat Rev Neurosci 2015;16:109–120.
3. Fagan AM, Minton MA, Mach RH, et al. Inverse relation in vivo amyloid imaging load and cerebrospinal fluid Abeta42 in humans. Ann Neurol 2006;59:512–519.
4. Tapiola T, Overmyer M, Lehtovirta M, et al. The level of cerebrospinal fluid tau correlates with neurofibrillary tangles in Alzheimer’s disease. Neuroreport 1997;8:3961–3963.
5. Tokuda T, Salem SA, Allopp D, et al. Decreased alpha-synuclein in cerebrospinal fluid of aged individuals and subjects with Parkinson’s disease. Biochem Biophys Res Commun 2006;349:162–166.
6. Mollenhauer B, Locascio JJ, Schule-Schaeffer W, Sixel-Doring F, Trenkwalder C, Schlossmacher MG. Alpha-Synuclein and tau concentrations in cerebrospinal fluid of patients presenting with parkinsonism: a cohort study. Lancet Neurol 2011;10:230–240.
7. Hong Z, Shi M, Chung KA, et al. DJ-1 and alpha-synuclein in human cerebrospinal fluid as biomarkers of Parkinson’s disease. Brain 2010;133:713–726.
8. Kang JH, Irwin DJ, Chen-Plotkin AS, et al. Association of cerebrospinal fluid beta-amyloid 1–42, T-tau, P-tau181, and alpha-synuclein levels with clinical features of drug-
naive patients with early Parkinson disease. JAMA Neurol 2013;70:1277–1287.

9. Kang JH, Mollenhauer B, Coffey CS, et al. CSF biomarkers associated with disease heterogeneity in early Parkinson’s disease: the Parkinson’s Progression Markers Initiative study. Acta Neuropathologica 2016;131:935–949.

10. Majbour NK, Vaikath NN, Eusebi P, et al. Longitudinal changes in CSF alpha-synuclein species reflect Parkinson’s disease progression. Mov Disord 2016;31:1535–1542.

11. Hall S, Ohrfelt A, Constantinescu R, et al. Accuracy of a panel of 5 cerebrospinal fluid biomarkers in the differential diagnosis of patients with dementia and/or Parkinson’s disorder. Arch Neurol 2012;69:1445–1452.

12. Stewart T, Liu C, Gintingha C, et al. Cerebrospinal fluid alpha-synuclein predicts cognitive decline in Parkinson disease progression in the DATATOP cohort. Am J Pathol 2014;184:966–975.

13. Zhang J, Mattison HA, Liu C, et al. Longitudinal assessment of tau and amyloid beta in cerebrospinal fluid of Parkinson disease. Acta Neuropathologica 2013;126:671–682.

14. Mollenhauer B, Zimmermann J, Snell-Doring F, et al. Monitoring of 30 marker candidates in early Parkinson disease as progression markers. Neurology 2016;87:168–177.

15. Gomperts SN, Locascio JJ, Makaretz SJ, et al. Tau positron emission tomographic imaging in the Lewy body diseases. JAMA Neurol 2016;73:1334–1341.

16. Irwin DJ, Grossman M, Weintraub D, et al. Neuropathological and genetic correlates of survival and dementia onset in synucleinopathies: a retrospective analysis. Lancet Neurol 2017;16:55–65.

17. The Parkinson Progression Marker Initiative (PPMI). Prog Neurobiol 2011;95:629–635.

18. Goetz CG, Tilley BC, Shaftman SR, et al. Movement Disorder Society–sponsored revision of the Unified Parkinson’s Disease Rating Scale (MDS-UPDRS): scale presentation and clinimetric testing results. Mov Disord 2008;23:2129–2170.

19. Tomlinson CL, Stone R, Patel S, Rick C, Gray R, Clarke CE. Systematic review of levodopa dose equivalency reporting in Parkinson’s disease. Mov Disord 2010;25:2649–2653.

20. Stiansy-Kolster K, Mayer G, Schafer S, Moller JC, Heinzel-Gutenbrunner M, Oertel WH. The REM sleep behavior disorder screening questionnaire: a new diagnostic instrument. Mov Disord 2007;22:2386–2393.

21. Nalls MA, Keller MF, Hernandez DG, et al. Baseline genetic associations in the Parkinson’s Progression Markers Initiative (PPMI). Mov Disord 2016;31:79–85.

22. Wang Y, Shi M, Chung KA, et al. Phosphorylated alpha-synuclein in Parkinson’s disease. Sci Transl Med 2012;4:121ra120.

23. Hansson O, Hall S, Ohrfelt A, et al. Levels of cerebrospinal fluid alpha-synuclein oligomers are increased in Parkinson’s disease with dementia and dementia with Lewy bodies compared to Alzheimer’s disease. Alzheimers Res Ther 2014;6:25.

24. Shahnawaz M, Tokuda T, Waragai M, et al. Development of a biochemical diagnosis of Parkinson disease by detection of alpha-synuclein misfolded aggregates in cerebrospinal fluid. JAMA Neurol 2017;74:163–172.

25. Fagan AM, Xiong C, Jasielec MS, et al. Longitudinal change in CSF biomarkers in autosomal-dominant Alzheimer’s disease. Sci Transl Med 2014;6:226ra230.

26. Bateman RJ, Xiong C, BENZINGER TL, et al. Clinical and biomarker changes in dominantly inherited Alzheimer’s disease. N Engl J Med 2012;367:795–804.

27. Schrag A, Horstfall L, Walters K, Noyce A, Petersen I. Prediagnostic presentations of Parkinson’s disease in primary care: a case-control study. Lancet Neurol 2015;14:57–64.

28. Ferreihenejad SM, Romenets SR, Anang JB, Lareille V, Gagnon JF, Postuma RB. New clinical subtypes of Parkinson disease and their longitudinal progression: a prospective cohort comparison with other phenotypes. JAMA Neurol 2015;72:863–873.

29. Vendette M, Gagnon JF, Decary A, et al. REM sleep behavior disorder predicts cognitive impairment in Parkinson disease without dementia. Neurology 2007;69:1845–1849.

30. Toledo JB, Vanderstichele H, Figuero M, et al. Factors affecting Abeta plasma levels and their utility as biomarkers in ADNI. Acta Neuropathologica 2011;122:401–413.

31. Schrag A, Siddiqui UF, Anastasius Z, Weintraub D, Schott JM. Clinical variables and biomarkers in prediction of cognitive impairment in patients with newly diagnosed Parkinson’s disease: a cohort study. Lancet Neurol 2017;16:66–75.

32. Mollenhauer B, Cullen V, Kahn I, et al. Direct quantification of CSF alpha-synuclein by ELISA and first cross-sectional study in patients with neurodegeneration. Exp Neurol 2008;213:315–325.

33. Mignini F, Bronzi et, Felici L, et al. Dopamine receptor immunohistochemistry in the rat choroid plexus. J Auton Pharmacol 2000;20:325–332.

34. Chau KY, Cooper JM, Schapira AH. Pramipexole reduces phosphorylation of alpha-synuclein at serine-129. J Mol Neurosci 2013;51:573–580.

35. Yu X, Yao JY, He J, Tian JW. Potentiation of rostatin-induced neuroinflammation and neurodegeneration by rotigotine-loaded microspheres. Life Sci 2015;124:136–143.

36. Tokuda T, Qureshi MM, Andah MT, et al. Detection of elevated levels of alpha-synuclein oligomers in CSF from patients with Parkinson disease. Neurology 2010;75:1766–1772.
Longitudinal CSF biomarkers in patients with early Parkinson disease and healthy controls

Brit Mollenhauer, Chelsea J. Caspell-Garcia, Christopher S. Coffey, et al.

Neurology 2017;89;1959-1969 Published Online before print October 13, 2017
DOI 10.1212/WNL.0000000000004609

This information is current as of October 13, 2017
