1 **Plasma neurofilament light and p-tau181 and risk of psychosis in Parkinson's disease**

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Abstract

Background: Neuropsychiatric symptoms are common and important to people with Parkinson's disease (PD) but their etiology is poorly understood. Plasma neurofilament light (NfL) and p-tau181 are biomarkers of neuro-axonal degeneration and tau pathology respectively which have yet to be explored in association with the affective and psychotic symptoms in PD.

Objective: To investigate the relationship between plasma NfL and p-tau181 with the affective and psychotic symptoms in PD.

Methods: We assessed the baseline concentration of plasma NfL and p-tau181 in a cohort of 108 patients with PD and 38 healthy controls. A subgroup of patients (n=63) were assessed annually with clinical measures for up to 7 years. Psychotic symptoms were assessed using Non-Motor Symptom Scale (NMSS) with affective symptoms measured in the Hospital Anxiety and Depression Scale (HADS).

Results: Baseline plasma NfL was a significant predictor of psychotic symptoms longitudinally across the study (adjusted for age, Hoehn and Yahr stage (HY), duration of follow up, duration of disease, dopaminergic medication and baseline cognition: (OR 6.23 [95% CI 1.30-29.8], p = 0.022) and was associated with shorter time to develop psychosis (adjusted R^2=0.20, p = 0.01). There was no association between NfL concentration and the cumulative prevalence of affective symptoms. Plasma p-tau181 concentration was not associated with psychotic or affective symptoms.

Conclusion: These findings suggest psychotic symptoms are associated with greater neurodegeneration in PD. No association was seen between plasma p-tau181 and these neuropsychiatric symptoms. Further studies are needed to explore NfL as a potential biomarker for psychosis in PD.

Introduction

Neuropsychiatric symptoms (NPS) such as anxiety, depression, psychosis and apathy are common in Parkinson's disease (PD) with almost all patients affected at some point during the disease course (1). Of these NPS, affective and psychotic symptoms are two of the most important determinants of quality of life in PD (2, 3). Affective and psychotic symptoms are a significant burden for people with PD, associated with earlier mortality (4), greater caregiver strain (5, 6) and earlier nursing home placement (7). While these symptoms can co-exist in PD, with negative consequences for cognitive performance (8), factor analyses support their existence as separate subsyndromes (9). Indeed, their respective clinical implications are hugely different; the treatment of PD psychosis (PDP) is particularly challenging due to the limited treatment options and increased risk in mortality associated with the use of antipsychotics (10). Furthermore, evidence suggests key differences in their mechanistic underpinnings with psychosocial determinants likely to contribute more significantly to affective symptoms (11).

Little is known about the biological mechanisms underlying the affective and psychotic symptoms in PD. Older age, cognitive impairment and longer duration of disease have been identified as predictive factors for both emergent psychosis and depression in PD, but no objective biomarkers exist and the underlying neurobiology remains poorly understood (12). However, both affective and psychotic symptoms can be seen in the prodromal phase of PD indicating that the neuropathological substrates of the PD contribute to their etiology (13, 14). Indeed, the affective and psychotic symptoms have been linked with widespread neurodegeneration both inside and outside nigrostriatal dopaminergic pathways in PD (15-18). In PDP, Alzheimer's disease (AD) pathology may also contribute, with post-mortem studies finding increased neurofibrillary tangles and greater burden of hyperphosphorylated

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103   tau associated with psychotic symptoms (19-21). The earlier emergence of psychotic

104   symptoms in Lewy body dementias relative to AD indicates alpha-synuclein may also be a

105   key contributor to the risk of psychosis, but this has not yet been clearly demonstrated in

106   studies (22). However, accumulation of alpha-synuclein in the nucleus accumbens, ventral

107   tegmental area and substantia nigra has been associated with depression in PD (17, 23).

108

109   The differing etiologies of the affective and psychotic symptoms in PD are likely

110   multifactorial but the relative importance of transmitter changes, neurodegeneration,

111   neuropathology and dopaminergic and other medications remains unclear (12, 24, 25). Better

112   characterization of the mechanisms behind these important symptoms is crucial to aid novel

113   drug discovery; identifying biomarkers for these NPS would help to determine their

114   biological correlates and, importantly, could offer prognostic value for at risk patients which

115   would lead to more careful monitoring and earlier management (26, 27).

116

117   Neurofilament light (NfL) is a specific biomarker for neuro-axonal damage, irrespective of

118   the underlying cause (28). Plasma and CSF NfL concentrations correlate closely, adding to its

119   promise as a potential candidate for use in clinical practice (29). Growing consensus suggests

120   that NfL may not be increased in the early stages of PD (30, 31), but higher NfL

121   concentrations are associated with faster disease progression and greater motor and cognitive

122   impairments (32, 33). Coexistent AD pathology is a common feature of dementia in PD

123   (PDD), seen in over 50% of cases post-mortem (34). Phosphorylated tau at threonine 181 (p-

124   tau181) is a newly established plasma biomarker, specific for AD tau pathology and

125   correlates closely with amyloid β pathology (35). Plasma p-tau181 has recently been shown

126   to predict cognitive decline in patients with dementia with Lewy bodies (DLB) (36).

127

128   However, to our knowledge no prior study has looked to investigate an association between

129   plasma NfL or p-tau181 and the affective or psychotic symptoms (A/PS) in PD. Any increase

130   in NfL in patients reporting A/PS would suggest neuro-axonal degeneration as a key factor in

131   the etiology and could offer prognostic value, while an increase in p-tau would indicate a

132   contribution of AD pathology to these symptoms. Here, the aim was to explore the

133   relationship between NfL and p-tau181 concentration with the cumulative prevalence of

134   A/PS. Given NPS are known to fluctuate in neurodegenerative disease (37), the cumulative

135   prevalence of these symptoms over time is more likely to reflect underlying neurobiological

136   changes measured by plasma biomarkers. A preferential increase in p-tau181 was expected

137   in patients with PDP reflecting the contribution of tau to the etiology suggested by post-

138   mortem studies, and greater concentrations of NfL was anticipated for both A/PS reflecting

139   the contribution of neurodegeneration to the neurobiology of these symptoms.

140

141   **Methods**

142   *Patient cohort*

143   Plasma samples were taken at study entry between 2012 to 2015 from 108 patients with a

144   diagnosis of probable idiopathic PD from the King's College Hospital center of the Non-

145   motor International Longitudinal Study (NILS) and 38 age- and sex-matched healthy controls

146   (HC). NILS is a cohort study designed to assess outcomes from non-motor symptoms in PD

147   over time, patients are assessed at baseline with clinical measures and plasma collection and a

148   subgroup (n=63, 58%) were followed up annually with clinical measures for up to 7 years

149   after inclusion (38). Follow up length was variable between patients with a median duration

150   of follow up of 4 years. Inclusion required a diagnosis of PD made by a neurologist according

151   to internationally recognized diagnostic criteria (39). Exclusion criteria include insufficient

152   plasma or clinical information, inability to give informed consent, clinical diagnosis of

153   dementia at baseline or atypical parkinsonism. Plasma samples from HC were retrieved from

154   the NIHR South London and Maudsley BioResource Centre. The NILS study was authorized

155   by local ethics committees (NRES Southeast London REC, 10084, 10/H0808/141). All

156   patients gave written consent prior to study procedures and all patient data were anonymized

157   and coded.

158

159   *Clinical data*

160   Data extracted for PD patients included sex, age, duration of disease (years), education

161   (years), follow up (FU) duration, dopaminergic medication history including levodopa

162   equivalent daily dose (LEDD) where available, Hoehn and Yahr stage (H&Y) (40), Scales for

163   Outcomes in Parkinson's Disease (SCOPA-motor) (41), Non-Motor Symptom Scale (NMSS)

164   (42), Hospital Anxiety and Depression Scale (HADS) (43) and Mini Mental State

165   Examination (MMSE) score (44).

166

167   The NMSS is a clinician-rated scale used in PD to assess the severity (0-3) and frequency (1-

168   4) of non-motor symptoms including illusions, hallucinations and delusions. Severity and

169   frequency are multiplied to give the total score for each item and a binary classification of

170   psychosis was applied for patients scoring ≥ 1 in either hallucinations or delusions. The

171   HADS is self-administered with patients meeting criteria for affective symptoms if they

172   scored ≥ 7 on the anxiety or depression items (45). Anxiety and depression were grouped

173   together as affective symptoms due to the commonality in their underlying etiology.

174   Cognitive impairment was clinician rated using the MMSE.

175

176   *Plasma NfL concentration 113*

177   Plasma NfL concentration was measured using the Simoa NF-Light Advantage kit on an HD-

178   X assay platform in n=143 samples (PD n=105, HC n=38) at the UK Dementia Research

179   Institute, University College London, UK. All plasma samples collected during the course of

180   the longitudinal cohort study were stored at −80°C until assayed. Testers were blinded to

181   samples and 91% (n = 130) were measured in duplicate (insufficient sample available for

182   n=13). Intra-assay coefficient of variation was 4.55% and inter-assay coefficient of variation

183   were 8.53% and 2.35% respectively for high and low controls. The limit of detection (LOD)

184   was 0.038 pg/mL and the lower limit of quantification (LLOQ) was 0.174 pg/mL.

185

186   *Plasma p-tau181 concentration*

187   Plasma p-tau was measured for 104 PD samples at King's College London using the

188   commercially available Simoa® pTau-181 V2 Advantage Kit (Quanterix; 103714). Plasma

189   was diluted 1:4 and read on the HD-1 analyzer. Data acquisition spanned 5 analytical runs,

190   the lower limit of quantification (LLOQ) for this assay was 0.127 pg/mL and the coefficient

191   variation (CV) for inter- and intra-assay variability was 7.51% and 7.69% respectively.

192

193   *Statistical analysis*

194   PD patients were grouped by A/PS at baseline and by cumulative prevalence of A/PS over

195   the duration of the study. For the longitudinal analysis, cases without FU were excluded as it

196   could not be determined if they developed A/PS in the unstudied period. Cumulative

197   prevalence includes cases with A/PS at baseline or emergent A/PS in FU. Cumulative

198   prevalence was used as the primary outcome given fluctuations in NPS are common in

199   neurodegenerative disease and so point prevalence would likely lead to underestimates of

200   symptomology (37). 'New cases' describe those followed longitudinally who developed

201   emergent A/PS in FU. Cases were symptoms were present at baseline but not during the

202   follow up period were included in the cumulative prevalence analysis but excluded in a

203   secondary analysis including only cases with persistent A/PS over the follow up period. Time

204   to A/PS was also calculated, for cases who did not develop A/PS within the study duration

205   the total time the individual was followed up for was imputed.

206

207   Across groups continuous variables were compared using the independent t-tests or Mann-

208   Whitney U-tests, distribution dependent. Categorical data were analyzed with the Chi squared

209   tests. The relationship of plasma NfL to age and gender were compared across PD and HC

210   groups.

211

212   Within PD patients, correlations between NfL, p-tau181 and baseline clinical outcomes were

213   assessed with Spearman's rank correlation. NfL and p-tau181 concentration were log-

214   transformed to achieve a normal distribution with the assumption of normality assessed with

215   Shapiro-Wilk tests. The log10 transformed data were used in all further analysis. To assess

216   the predictive power of plasma NfL and p-tau181 concentration for A/PS, the cumulative

217   incidence of A/PS was used in logistic regression. These logistic regression analyses were

218   adjusted for age, baseline MMSE, H&Y stage, dopaminergic medication, duration of disease

219   and duration of follow up as these are well-established correlates and predictors of A/PS in

220   PD. LEDD was not available for all cases with longitudinal follow up and so use of

221   dopaminergic medication was used as a covariate. The relationship between time to develop

222   psychosis was explored with linear regression for both NfL and p-tau181 adjusted for age,

223   duration of disease, baseline MMSE, H&Y stage and LEDD. Linear regression was also used

224   to identify associations between NfL and p-tau181 with age, motor and cognitive outcomes.

225   Secondary logistic regression was performed for cases for whom A/PS persisted throughout

226   FU duration and for incident psychosis during FU.

227

228   Significance threshold was set to $p < 0.05$, where applicable values are given for two-tailed

229   tests. Statistical tests were carried out with Stata version 16.0.

230

231   **Results**

232   *Demographic and clinical data*

233   108 PD patients (26F; mean age 63.1±12.4 years) were included with 38 age- and sex-

234   matched HC (12F, 63.2±12.4). Sixty-three (58%) PD patients (16F; mean age 62.6±12.0)

235   were followed longitudinally for mean 3.69±1.76 years. At baseline, mean MMSE 28.6±2.59,

236   mean H&Y 2.33±0.76 and mean duration disease 6.61±5.92 years.

237

238   At baseline, 55 (51%) participants reported at least one A/PS, 23 (22%) with psychotic

239   symptoms (hallucinations n=21, delusions n=9) and 50 (46%) with affective symptoms

240   (depression n=32, anxiety n=35). *Table 1*. In the longitudinal follow up, 50 (74%) cases

241   reported A/PS, in 27 cases persistent from baseline, 19 new onset during follow up period

242   and 4 with symptoms at baseline which resolved during follow up period. In patients with

243   follow up, 29 had psychotic symptoms (hallucinations n=27, delusions n=17), 14 persistent

244   from the baseline assessment and 15 new onset during the follow up period. Forty-eight

245   (76%) reported affective symptoms (depression n=37, anxiety n=43), 23 persistent from

246   baseline, 21 emergent in the follow up period and 4 for whom affective symptoms at baseline

247   were no longer present during follow up. The evolution of neuropsychiatric symptoms during

248   the course of the study is illustrated in Table 1.

249

250   *Clinical characteristics of patients with psychotic and affective symptoms*

251   Demographic and clinical outcomes for PD patients with and without psychosis cumulative

252   throughout the study are presented in Table 2. Patients with psychotic symptoms displayed

253   more significant motor impairments (SCOPA-motor: U=-2.69, p = 0.007) and reported a

10

254 higher burden of affective symptoms (HADS: U =-2.55, p = 0.01) without evidence of more

255 advanced stage of disease (H&Y stage U = -0.40, p = 0.69). Levodopa equivalent daily dose

256 (LEDD) was significantly higher in those with psychotic symptoms than those without

257 (984mg v 472 mg, U=-3.16, p = 0.002). Baseline MMSE was equivalent across those with or

258 without psychosis but patients with psychosis had significantly greater annual decline in

259 MMSE (U = -2.23, p = 0.03). Duration of follow up was equivalent in those with or without

260 psychosis but patients with psychosis trended towards greater duration of disease (U = -1.88,

261 p = 0.06).

262

263 The majority of PD patients (76%) met criteria for anxiety or depression at some point in the

264 duration of the study. Demographic and clinical outcomes for affective symptoms are

265 included in Table 2. There was no difference in cognition or disease stage in patients with or

266 without affective symptoms but those with affective symptoms showed significantly greater

267 degree of motor impairment in the SCOPA-motor (18.4 v 10.9, U =-2.88, p = 0.001).

268

269 *Plasma NfL concentration and A/PS*

270 There was no difference in NfL levels between HC (mean 23.5pg/mL $\pm$20.2) and PD samples

271 (mean 26.2pg/mL $\pm$16.3), logNfL t(139) = -1.86, p = 0.06. In linear regression increased age

272 was predictive of greater plasma NfL levels R^2=0.17, p < 0.001. No gender differences were

273 seen in NfL concentrations logNfL t(139) = -0.62, p = 0.54.

274

275 In logistic regression adjusted for age, baseline MMSE, H&Y stage, dopaminergic

276 medication, duration of disease and duration of follow up, higher NfL concentration was a

277 significant predictor of psychosis across the duration of the study (OR 6.23 [95% CI 1.30-

278 29.8], p = 0.022) *Table 3*. Higher NfL concentration was also correlated with shorter time to

279   develop psychotic symptoms (r(68) =-0.40, p < 0.001) and in linear regression adjusted for

280   age, duration of disease, H&Y stage, LEDD and baseline MMSE, NfL was a significant

281   predictor of time to psychosis (adjusted R^2=0.20, p = 0.01). There was trend association

282   between NfL concentration and new onset psychosis in the follow up period adjusted for age,

283   baseline cognition, H&Y, duration of disease and duration of follow up (OR 5.47 [0.88-34.2],

284   p=0.069). However, NfL was not significantly associated with either hallucinations or

285   delusions alone in logistic regression adjusted for age, baseline MMSE, H&Y stage,

286   dopaminergic medication, duration of disease and duration of follow up (hallucinations OR

287   3.76 [0.84-16.8], p = 0.08; delusions OR 2.24 [0.46-11.02], p = 0.32).

288

289   There was no correlation between plasma NfL and HADS scores at baseline (rho = 0.04, p =

290   0.68). Cumulative affective symptoms across the duration of study was not associated with

291   higher baseline plasma NfL in logistic regression adjusted for age, baseline cognition, H&Y

292   stage, dopaminergic medication, duration of disease and duration of follow up (OR 3.41

293   [0.60-19.4], p = 0.18). *Table 3*. However, if analysis was restricted to those who had affective

294   symptoms during the follow up period ie excluding those with affective symptoms at baseline

295   which resolved during the FU period (n=4), then in a logistic regression model adjusted for

296   age, baseline MMSE, HY stage, dopaminergic medication, duration of disease and duration

297   of FU, baseline NfL became a significant predictor of affective symptoms (OR 14.4 [2.01-

298   104.5], p = 0.008).

299

300   In PD patients, NfL concentration was correlated with the duration with PD (r(105) = 0.27, p

301   = 0.01). Higher plasma NfL was correlated with lower baseline MMSE score (r(105) = -0.30,

302   p = 0.002).

303

304 Plasma NfL was positively correlated with scores on SCOPA-motor r(81) = 0.28, p = 0.012.

305 In linear regression adjusted for age, NfL was a significant predictor of SCOPA motor scores

306 adjusted R^2=0.23, p = 0.01. NfL was also positively correlated with H&Y (r(85) 0.27, p =

307 0.01). NfL concentration was higher in participants lost to follow up but the difference was

308 not significant logNfL t(103) = 1.72, p = 0.09.

309

310 *Plasma p-tau181 concentration*

311 Increased age was predictive of greater plasma p-tau181 levels in linear regression (Pearson

312 corr 0.18; R^2=0.02, p = 0.07). No sex differences in p-tau181 were seen across patients with

313 PD logp-tau(104) = 1.12, p = 0.26. Plasma p-tau was also higher in participants lost to follow

314 up but the difference was not significant logp-tau t(102) = 1.31, p = 0.19.

315

316 In logistic regression adjusted for age, H&Y, duration of disease, duration of FU,

317 dopaminergic medication and baseline MMSE, plasma p-tau181 showed no association with

318 psychosis in the study period (OR 5.17 (95% CI 0.38-70.0), p = 0.22). *Table 3*. There was no

319 correlation between baseline HADS score and plasma p-tau181 (r(102)=-0.05, p=0.59) and

320 there was no association between p-tau181 concentration and cumulative affective symptoms

321 across the course of the study in adjusted logistic regression (OR 0.17 [95% CI 0.01-3.22], p

322 = 0.24).

323

324 **Discussion**

325 In the first longitudinal study to explore the relationship between plasma NfL and p-tau181

326 with the affective and psychotic symptoms in PD, increased NfL concentration was

327 associated with both greater longitudinal risk of PDP and significantly shorter time to

328 psychosis. Plasma NfL concentration was not associated with the cumulative prevalence of

329   affective symptoms but was associated with greater odds of persistent affective symptoms.

330   We did not see any association between p-tau181 concentration and psychotic or affective

331   symptoms.

332

333   NfL is a well-established cross-disease biomarker of axonal degeneration (28). The higher

334   concentration of NfL in PDP suggests a role for neurodegeneration in the etiology of these

335   symptoms. H&Y stage was adjusted for in all models with equivalent staging in those with or

336   without psychosis making it an unlikely confounder. These findings are perhaps unsurprising

337   given PDP is associated with extensive neurodegeneration of limbic, paralimbic and

338   neocortical gray matter (18) and has recently been associated with increased density of Lewy

339   bodies and greater neuronal loss and gliosis both inside and outside the substantia nigra (20,

340   46). In patients with mild cognitive impairment, emergent mild behavioral impairment has

341   also been associated with increases in NfL suggestive that neurodegeneration may drive these

342   clinical symptoms at an early stage (47). PDP likely represents a complex intersection of

343   exogenous and endogenous factors with the current study emphasizing the likely contribution

344   of neurodegeneration in their etiology, this may have translational relevance for other

345   psychotic disorders given susceptibility to psychosis is increasingly viewed trans-

346   diagnostically (48). The trend association between incident psychosis and NfL suggests a

347   larger sample size is required to investigate the potential of NfL as a biomarker to indicate

348   patients at risk of future PDP who would benefit from more frequent monitoring and earlier

349   intervention. Furthermore, while hallucinations and delusions were not associated with NfL

350   concentration individually, this also likely reflects the overall smaller numbers with positive

351   symptomology. Future studies should aim to explore the relationship between NfL and

352   hallucinations and delusions separately.

353

354 Previous post-mortem studies have suggested AD pathology may also contribute to PDP with

355 hallucinations associated with a widespread increase in beta-amyloid plaque and tangle

356 densities in later stage PD (20). Our results did not reflect these post-mortem findings, which

357 perhaps reflects the earlier stage of PD of the patients included in the study, typically PDP at

358 the mild cognitive impairment stage has Lewy bodies mainly restricted to the amygdala with

359 limited AD pathology (49). Further studies with larger sample sizes, more advanced PD and

360 more comprehensive follow up are needed to explore the role of p-tau181 as a marker of

361 psychosis in PD.

362

363 No increase in NfL or p-tau181 was seen in patients with cumulative prevalence of affective

364 symptoms in the study. However, baseline NfL was a significant predictor of persistent

365 affective symptoms during the study, suggesting neurodegenerative processes may be more

366 prominent where affective symptoms are more established. However, only a minority of

367 patients (n = 15, 24%) did not report affective symptoms during the study and so it could be

368 that the study is underpowered to detect differences for the cumulative prevalence of

369 affective symptoms. An increase in NfL might have been expected for patients with

370 depression given the development of affective symptoms is particularly associated with

371 neuronal loss and gliosis in the locus coerulus and substantia nigra (16, 17, 46). However,

372 while in some cases depression likely develops due to pathological changes inherent to PD, in

373 other cases depression may be incidental or intrinsic to the comorbidity of a chronic

374 condition with greater psychosocial influences rather than specifically related to

375 neurodegenerative processes in PD (50). Where the etiology of depression differs, the

376 underlying neurobiology may also differ which could cause variation in the degree of

377 neurodegeneration and subsequent NfL increase in PD patients with depression.

378

379   *Limitations*

380

381   While the longitudinal nature of this study is one of its major strengths, the attrition rate and

382   variation in the length of follow up could affect the estimates of patient numbers developing

383   A/PS. To calculate the cumulative frequency of A/PS, patients without follow up in the study

384   were excluded which significantly reduced our sample size. This may underlie some of the

385   negative findings in the study and future studies should address this issue. Furthermore, NfL

386   and p-tau181 concentrations were only measured at baseline and so we were not able to

387   monitor changes in these biomarkers in patients who developed A/PS during the study, this

388   would have been particularly interesting given associations with NfL were seen with

389   cumulative incidence. Future studies should aim to investigate whether emergence of A/PS

390   over time are associated with further increases in NfL.

391

392   LEDD was not available for all cases in the study with longitudinal follow up (n=55).

393   Therefore a binary classification for the use of dopaminergic medication or not was used as a

394   proxy to adjust for this confounder in the logistic regression models. Given this does not give

395   information as to the dose of levodopa received this is a limitation of the study. However,

396   while LEDD was significantly associated with cumulative prevalence of psychosis, LEDD

397   was not correlated with NfL ($r = 0.14$, $p = 0.22$). LEDD was also included as a covariate in

398   regression models where NfL was significantly associated with shorter time to psychosis.

399   Thus while it will be important to include LEDD in future studies we do not feel that this

400   limitation undermines the findings of the current study.

401

402   A/PS were assessed in this study using the NMSS for psychotic symptoms and HADS for

403   affective symptoms. While the NMSS allows for the assessment of severity and frequency of

404   hallucinations and delusions and correlates closely with similar items on the Neuropsychiatric

405   Inventory (NPI), the perceptual and hallucinatory domains have lower internal consistency

406   (51). Furthermore, the NMSS lacks the breadth of the NPI and does not include a number of

407   symptoms known to be common to PD such as apathy and impulse control disorders (52).

408   We were therefore unable to adjust for the overall NPS burden or other potentially

409   overlapping symptoms such as agitation or apathy in our analysis. Other scales such as the

410   MDS-NMS offer greater phenomenological detail than the NMSS assessing illusions,

411   passage and presence hallucinations in greater detail (53). The HADS was used to

412   supplement the NMSS due to the greater diagnostic detail it offers for symptoms of

413   depression and anxiety. The HADS is validated for use in PD (54) and in our study was

414   closely correlated with the mood domain of the NMSS ($r=0.7$) but the use of differing scales

415   for the affective and psychotic symptoms in the study means they were not assessed

416   uniformly and may have led to overestimates of affective symptoms. Future studies should

417   aim to use additional measures of NPS which are validated in PD and have fine-grained

418   assessment of A/PS.

419

420   This longitudinal cohort study was designed with MMSE rather than the Montreal Cognitive

421   Assessment (MoCA) which is known to be more sensitive in PD (55). However, while the

422   MoCA shows greater variability in PD, MMSE has been shown to be a suitable scale to

423   measure cognitive abilities in PD and cognition was not the primary end point of this study,

424   use of this scale should not affect the validity of our results (55).

425

426   *Conclusions*

427   We demonstrate the potential of NfL as a predictive marker for the cumulative prevalence of

428   psychotic symptoms in PD. This not only points to axonal neurodegeneration as an important

429 etiological factor in the development of psychosis but also suggests future promise as a

430 prognostic marker for these common and hard to treat symptoms. Further studies are needed

431 to explore the longitudinal characterization of A/PS with a wide array of biomarkers, both

432 new and existing.

433

460 **Conflicts of interest**

461 HZ has served at scientific advisory boards and/or as a consultant for Abbvie, Alector,

462 Annexon, Artery Therapeutics, AZTherapies, CogRx, Denali, Eisai, Nervgen, Pinteon

463 Therapeutics, Red Abbey Labs, Passage Bio, Roche, Samumed, Siemens Healthineers,

464 Triplet Therapeutics, and Wave, has given lectures in symposia sponsored by Cellectricon,

465 Fujirebio, Alzecure, Biogen, and Roche, and is a co-founder of Brain Biomarker Solutions in

466 Gothenburg AB (BBS), which is a part of the GU Ventures Incubator Program (outside

467 submitted work).

468 DA has received research support and/or honoraria from Astra-Zeneca, H. Lundbeck,

469 Novartis Pharmaceuticals, Evonik, and GE Health and has served as paid consultant for H.

470 Lundbeck, Eisai, Heptares, Mentis Cura, Eli Lilly, and Biogen.

471 KRC is in the advisory board of AbbVie, UCB, GKC, Bial, Cynapsus, Lobsor, Stada,

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478

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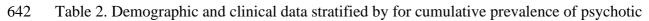527 2018;90(18):e1618-e26.

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636   Table 1. Cases by neuropsychiatric symptom across study duration. A/PS: affective or
637   psychotic symptom.
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	Baseline (n=108)		Follow up (n=63)	
	Cases	Evolution of symptoms from baseline	New cases	Total cases
Any A/PS	55 (51%)	27 persistent 4 resolved 24 no FU	19 (31%)	50 (80%)
Psychosis (hallucination or delusion)	23 (22%)	14 persistent 9 no FU	15 (24%)	29 (46%)
Hallucination	21 (20%)	9 persistent 5 symptoms resolved 7 no FU	13 (21%)	27 (43%)
Delusion	9 (8%)	5 persistent 4 no FU	12 (19%)	17 (27%)
Illusion or perceptual problem	13 (12%)	8 persistent 1 symptoms resolved 3 no FU	10 (16%)	19 (29%)
Affective	50 (46%)	23 persistent 4 resolved 23 no FU	21 (41%)	48 (76%)
Depression	32 (30%)	15 persistent 2 resolved 15 no FU	20 (33%)	37 (59%)
Anxiety	35 (32%)	15 persistent (47%) 3 symptoms resolved 17 no FU	25 (39%)	43 (69%)

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642   Table 2. Demographic and clinical data stratified by for cumulative prevalence of psychotic

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643 and affective symptoms in longitudinal follow up. H&Y: Hoehn and Yahr stage. LEDD:

644 levodopa equivalent daily dose. [*]n=55

	Psychosis+ (n=29)	Psychosis- (n=34)	Test statistic	p	Affective+ (n=48)	Affective- (n=15)	Test statistic	p
Age	63.7 (12.1)	61.7 (12.0)	$t(61)=-0.66$	0.51	61.9 (12.8)	64.9 (8.89)	$t(61)=0.84$	0.41
Female	31% (9)	21% (7)	$X^2=0.90$	0.34	25% (12)	27% (4)	$X^2=0.02$	0.90
Years of education	15.4 (4.84)	15.8 (4.44)	U=0.17	0.87	15.9 (4.64)	14.8 (4.51)	U=-0.75	0.45
Duration of disease (yrs)	6.10 (3.40)	4.63 (4.19)	U=-1.88	0.06	5.27 (3.83)	5.41 (4.19)	U=0.05	0.96
Baseline MMSE	28.3 (3.27)	29.3 (1.48)	U =1.59	0.11	28.6 (2.78)	29.4 (1.12)	U =0.85	0.39
Baseline HADS	11.9 (7.73)	7.62 (5.38)	U =-2.69	**0.007**	11.3 (6.97)	4.07 (1.58)	U =-4.46	**<0.001**
Baseline H&Y stage	2.38 (0.73)	2.32 (0.77)	U = -0.40	0.69	2.35 (0.73)	2.33 (0.82)	U =-0.13	0.90
Baseline SCOPA-motor	19.1 (6.50)	14.6 (9.34)	U =-2.50	**0.01**	18.4 (8.26)	10.9 (6.11)	U =-3.24	**0.001**
LEDD[*] (mg)	984 (721)	472 (439)	U=-3.16	**0.002**	753 (663)	508 (472)	U =-1.10	0.27
Duration of follow up (years)	3.72 (1.78)	3.67 (1.78)	U=0	1.00	3.72 (1.79)	3.40 (1.71)	U =-0.37	0.71
Annual MMSE decline	0.88 (1.51)	0.37 (0.88)	U = -2.23	**0.03**	0.65 (1.26)	0.48 (1.17)	U =-1.08	0.28

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657   Table 3. Baseline plasma NfL and p-tau181 concentration stratified by neuropsychiatric

658   symptoms in logistic regression models adjusted for age, baseline MMSE, duration of follow

659   up, duration of disease, dopaminergic medication and H&Y stage.

	Mean plasma NfL concentration (pg/mL) (mean+SD) n=63				Mean plasma p-tau concentration (pg/mL) (mean+SD) n=63			
	Symptom present	Symptom absent	OR [95% CI]	p	Symptom present	Symptom absent	OR [95% CI]	p
Affective symptoms	23.9+12.4	21.4+8.99	3.41 [0.60-19.4]	0.17	2.11+1.12	2.64+1.97	0.17 [0.01-3.23]	0.24
Depression	23.9+12.0	22.5+11.2	3.27 [0.70-15.4]	0.13	2.14+1.18	2.40+1.65	0.32 [0.02-4.05]	0.38
Anxiety	24.5+12.7	20.6+8.53	5.62 [0.98-32.1]	0.05	2.18+1.13	2.36+1.80	0.90 (0.07-11.4)	0.94
Psychosis (hallucinations +/- delusions)	26.1+11.4	20.9+11.5	6.23 [1.30-29.8]	**0.022**	2.43+1.10	2.07+1.54	5.17 [0.38-70.0]	0.22
Hallucination	26.2+11.7	21.1+11.2	3.76 [0.84-16.8]	0.08	2.43+1.12	2.08+1.52	4.11 [0.32-52.2]	0.28
Delusion	24.4+9.84	22.9+12.3	2.24 [0.46-11.0]	0.32	2.28+1.00	2.22+1.48	1.51 [0.09-25.7]	0.78
Illusions and perceptual difficulties	23.2+11.3	23.3+11.9	0.78 [0.16-3.89]	0.76	2.16+1.24	2.27+1.42	1.35 [0.11-16.8]	0.81

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