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BIOMARKERS

POSTER PRESENTATION

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BIOMARKERS (NON-NEUROIMAGING)

Associations between CSF complement factors and biomarkers of amyloid, tau, neurofilament light chain, and α-synuclein in AD, DLB, and PD

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Abstract

Background: While evidence suggests complement system involvement in Alzheimer's disease (AD), dementia with Lewy bodies (DLB), and Parkinson's disease (PD), its association with disease biomarkers remains unclear. We investigated the relationship of complement factors with amyloid, tau, NfL, and α-synuclein in CSF in AD, DLB, PD, and controls.

Method: We included 321 individuals with AD, DLB, PD, and controls from 6 centers of the EPND study. CSF Aβ42/40, *p*-tau181, NfL, and α-syn were centrally measured using NeuroToolKit (Roche Diagnostics), and 14 CSF complement factors using Milliplex (Merck KGaA). Controls were defined as normal cognition and normal Aβ42/40, whereas AD as abnormal Aβ42/40 without meeting clinical criteria of DLB or PD. Linear regression models adjusted for age and sex were used. Associations were post-hoc compared between individuals with low($\leq$23), intermediate(24-27), and high($\geq$28) MMSE scores.

Result: Sample characteristics are presented in Table 1. Lower Aβ42/40 levels were associated with lower levels of 7 complement factors in controls and with higher C1q and C2 levels specifically in AD (Figure 1, Figure 2). No associations of Aβ42/40 with complement were found in DLB and PD. Higher *p*-tau181 levels were associated with increased levels of 7 complement factors in controls and 6 in AD, and showed fewer associations in DLB and PD. The strength of *p*-tau181 associations with complement was similar across groups. Higher NfL levels were widely associated with higher complement factor levels in controls (13) and AD (12), and less in PD (6) and DLB (4). Higher α-syn levels were broadly associated with higher complement factor levels in

AD (13), controls (12), and DLB (12), but only minimally in PD (1). The strength of these NfL and α-syn associations with complement was not disease-specific. Conversely, compared to all groups, in PD higher α-syn levels were associated with lower C5, C5a, C9, factor-I and properdin levels. Individuals with intermediate MMSE scores largely drove the associations of α-syn with complement in AD. MMSE level did not clearly impact other associations.

Conclusion: CSF complement factors were associated with amyloid, tau, NfL, and α-synuclein, suggesting complement system involvement in several neurodegenerative diseases. Complement showed disease-specific associations with amyloid in AD and α-synuclein in PD.

Table 1. **Sample characteristics.**

	Control	AD	DLB	PD	Total sample
N	47	147	53	74	321
Age	63.7 (9.6) [B,C]	69.3 (8.3) [A,D]	69.6 (6.7) [A,D]	62.7 (10.2) [B,C]	67.0 (9.2)
Female sex (%)	14 (30%)	66 (45%) [C]	9 (17%) [B]	24 (32%)	113 (35%)
Education years	14.6 (3.4) [B,C,D]	12.8 (3.4) [A]	11.9 (2.8) [A,D]	13.2 (3.5) [A,C]	13.0 (3.4)
***APOE*-ε4 carriers (%)**	4 (8.7%) [B,C]	94 (64%) [A,D]	24 (51%) [A,D]	16 (25%) [B,C]	138 (45%)
MMSE score	29.0 (1.1) [B,C]	25.2 (4.0) [A,C,D]	23.8 (3.7) [A,B,D]	28.5 (1.7) [B,C]	26.3 (3.7)
MDS-UPDRS-III score	2.9 (2.6) [C,D]	6.0 (7.9) [C,D]	19.8 (15.2) [B,D]	31.3 (15.0) [A,B,C]	25.5 (16.9)
Aβ42/40	0.07 (0.01) [B,C]	0.03 (0.01) [A,C,D]	0.05 (0.02) [A,B,D]	0.07 (0.01) [B,C]	0.05 (0.02)
P-tau181	16.2 (5.3) [B,C]	28.6 (13.5) [A,C,D]	19.5 (6.5) [A,B,D]	15.5 (5.7) [B,C]	22.3 (11.7)
NfL	116.7 (81.3) [B,C]	197.3 (189.3) [A,D]	212.9 (232.1) [A,D]	145.9 (168.7) [B,C]	176.2 (183.6)
α-Syn	153.0 (324.1)	145.8 (257.4) [D]	121.4 (103.7)	112.3 (67.9) [B]	135.1 (220.0)
C1q	211.1 (88.1)	213.8 (141.6)	215.8 (94.7)	205.5 (75.3)	211.8 (114.1)
C2	132.2 (95.9) [C]	130.6 (72.3) [C]	177.9 (78.8) [A,B,D]	110.4 (45.7) [C]	134.0 (75.1)
C3	4110.3 (2545.0)	3681.8 (2344.1) [C]	5170.2 (3870.0) [B]	4251.7 (3035.2)	4121.7 (2872.0)
C3b	557.7 (710.6) [B]	378.2 (507.2) [A,C,D]	805.8 (1318.2) [B]	680.8 (1079.5) [B]	544.8 (875.5)
C4	1663.7 (731.5)	1553.1 (908.7)	1577.1 (738.3)	1584.0 (756.9)	1580.4 (821.1)
C4b	136.3 (82.1)	135.4 (74.8)	150.1 (77.8)	134.5 (81.5)	137.8 (77.8)
C5	52.3 (63.5)	48.6 (94.1) [C,D]	63.5 (74.5) [B]	52.2 (50.1) [B]	52.4 (78.2)
C5a	91.9 (95.5)	80.2 (104.9) [C,D]	117.7 (121.3) [B]	102.4 (91.6) [B]	93.2 (104.1)
C9	93.4 (90.9)	83.8 (75.4) [C,D]	110.9 (69.8) [B]	110.1 (89.4) [B]	95.7 (80.9)
Factor D	29.5 (14.2)	27.9 (13.2) [C]	32.6 (11.3) [B]	29.5 (13.8)	29.3 (13.2)
Factor I	200.5 (134.5)	172.4 (116.9) [C,D]	194.1 (85.8) [B]	195.3 (79.7) [B]	185.4 (107.8)
Factor H	671.6 (352.0)	609.7 (460.2)	662.6 (339.4)	640.1 (257.0)	634.5 (385.8)
Factor B	629.1 (409.9) [B]	493.0 (367.1) [A,D]	545.5 (269.0)	567.5 (305.0) [B]	538.8 (347.8)
Properdin	7.5 (9.3)	8.4 (30.6) [C,D]	8.5 (11.5) [B]	7.0 (4.3) [B]	8.0 (21.6)

The table shows key characteristics of the overall group and of each diagnostic group. Values represent mean (SD) for continuous variables, or N (%) for dichotomous variables. Education years were available for N=314 individuals, APOE-ε4 carriership for N=304, MMSE score for N=310, and MDS-UPDRS-III score for N=104. CSF Aβ42/40, p-tau181, NfL, and α-syn levels are expressed in pg/mL, whereas CSF complement factors levels in ng/mL. Differences between groups were tested with ANOVA for continuous variables and Chi-Squared for dichotomous variables. For analyses, all CSF measures were log-transformed, and their outliers were winsorized at ± 3 SD from mean to improve model fits. Unadjusted group comparisons $p<0.05$ compared to: (A) controls, (B) AD, (C) DLB, (D) PD. CSF Aβ42/40, p-tau181, NfL, and α-syn were measured via the NeuroToolKit, a panel of exploratory prototype assays designed to robustly evaluate biomarkers associated with key pathologic events characteristic of AD and other neurological disorders, used for research purposes only and not approved for clinical use (Roche Diagnostics International Ltd, Rotkreuz, Switzerland); all other product names and trademarks are the property of their respective owners. Abbreviations: APOE = Apolipoprotein E, MDS-UPDRS-III = Movement Disorders Society Unified Parkinson Disease Rating Scale Part III.

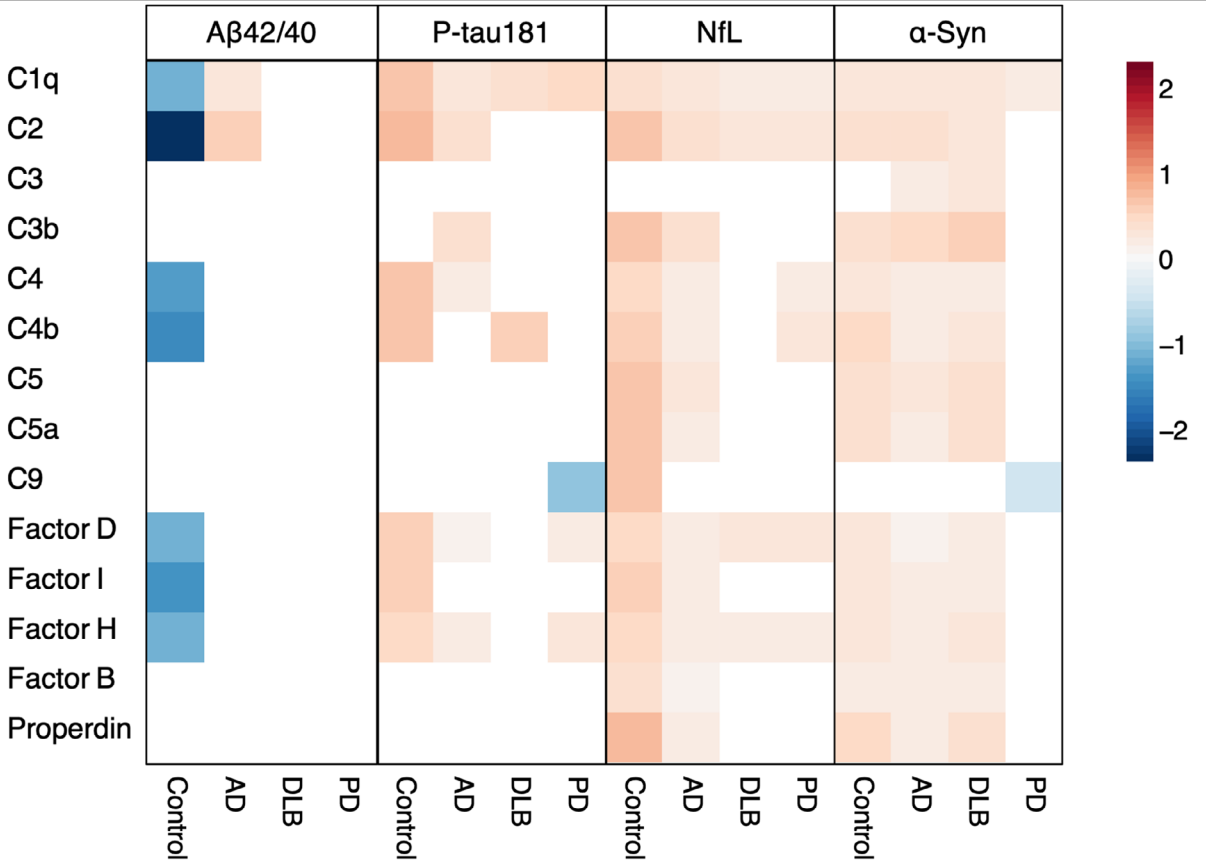

***Figure 1*. Associations of complement factors with ATN and α-synuclein markers in CSF in AD, DLB, PD, and controls.** The heatmap shows linear associations between levels of complement factors and levels of Aβ42/40, p-tau181, NfL, and α-syn in CSF in AD, DLB, PD, and controls. Red colours represent higher complement factor levels with more abnormal Aβ42/40, p-tau181, and NfL levels, and with higher α-syn levels (positive associations), whereas blue colours represent associations of an opposite direction. White represents lack of statistically significant associations ($p \geq 0.05$). For analyses, all CSF measures were log-transformed, and their outliers were winsorized at ± 3 SD from mean to improve model fits.

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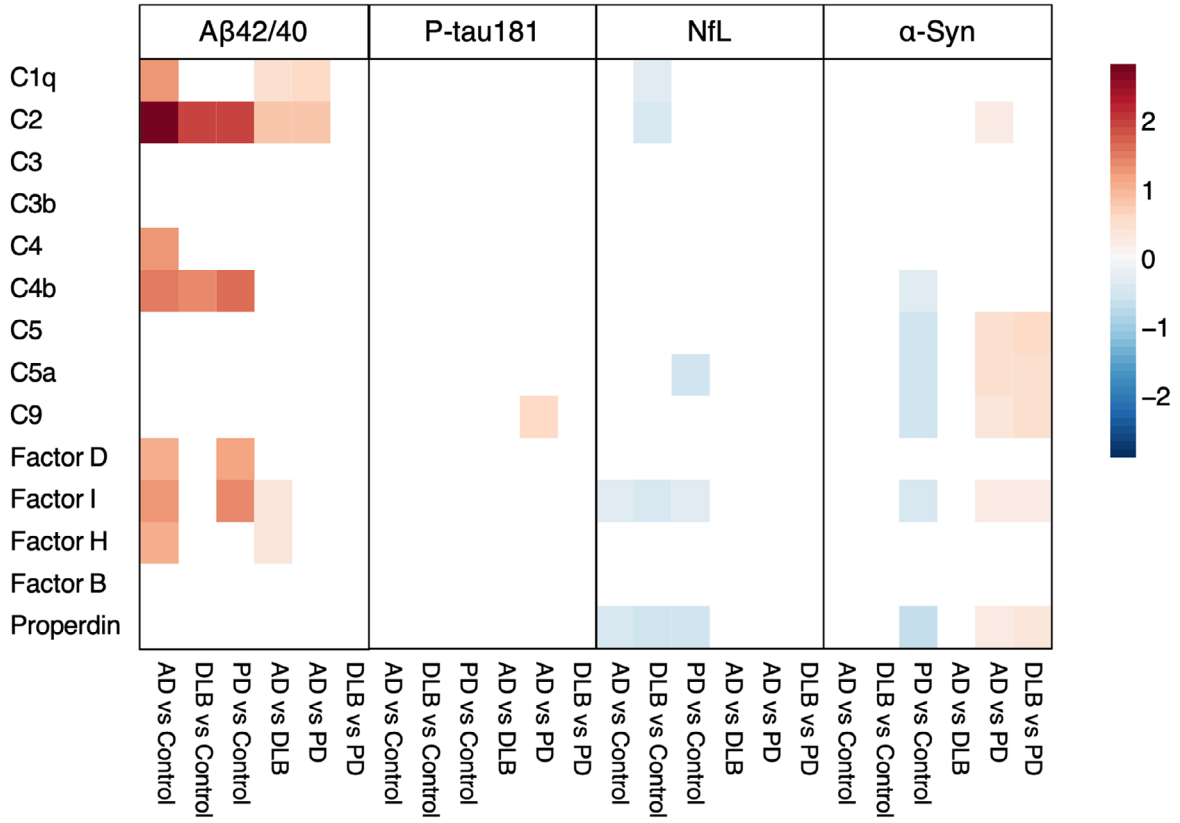

Figure 2. **Differences between AD, DLB, PD, and controls in the associations of complement factors with ATN and α-synuclein markers in CSF.** The heatmap shows the differences between AD, DLB, PD, and controls in the associations between levels of complement factors and levels of Aβ42/40, p-tau181, NfL, and α-syn in CSF. Red colours represent a more positive association (i.e., higher complement factor levels with more abnormal Aβ42/40, p-tau181, and NfL levels, and with higher α-syn levels) in the first *versus* the second group, whereas blue colours represent a more negative association in the first *versus* the second group. White represents lack of statistically significant differences between the two groups in the associations ($p \geq 0.05$). For analyses, all CSF measures were log-transformed, and their outliers were winsorized at ± 3 SD from mean to improve model fits.