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Neuroinflammation modulates the association of PGRN with cerebral amyloid-β burden

Wei Xu[a,*], Chen-Chen Tan[a], Xi-Peng Cao[b], Lan Tan[a,*], for the Alzheimer's Disease Neuroimaging Initiative[1]

[a] Department of Neurology, Qingdao Municipal Hospital, Qingdao University, Qingdao, China
[b] Clinical Research Center, Qingdao Municipal Hospital, Qingdao, China

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ABSTRACT

Progranulin (PGRN) and neuroinflammatory markers increased over the course of Alzheimer's disease (AD). We aimed to determine whether neuroinflammation could modulate the association of PGRN with amyloid pathologies. Baseline cerebrospinal fluid (CSF) PGRN and AD pathologies were measured for 965 participants, among whom 228 had measurements of CSF neuroinflammatory markers. Causal mediation analyses with 10,000 bootstrapped iterations were conducted to explore the mediation effects within the framework of A/T/N biomarker profile. Increased levels of CSF PGRN and inflammatory markers (sTNFR1, sTNFR2, TGF-β1, ICAM1, and VCAM1) were associated with T- or N-positive (TN+) profile, irrespective of the amyloid pathology. In TN+ group, CSF PGRN was associated with increased levels of these inflammatory markers and CSF amyloid-$\beta_{1\text{-}42}$ ($p < 0.01$). The neuroinflammatory markers significantly modulated (proportion: 20%~60%) the relationship of amyloid burden with CSF PGRN, which could predict slower cognitive decline and lower AD risk in the TN+ group. The abovementioned associations became non-significant in the TN- group. These findings revealed a close relationship between neuroinflammation and CSF PGRN in contributing to AD pathogenesis, and also highlighted the specific roles of PGRN in neurodegenerative conditions. Future experiments are warranted to verify the causal relationship.

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1. Background

Progranulin (PGRN) is a secreted glycoprotein ubiquitously expressed in peripheral organs and central nervous system (CNS). Its deficiency was associated with neuroinflammation (Ma et al., 2017; Takahashi et al., 2017) and neurodegenerative diseases (Götzl et al., 2018; Paushter et al., 2018) such as Alzheimer's disease (AD) (Minami et al., 2014; Xu et al., 2017; Xu et al., 2020). However, little is known about the biological mechanisms by which PGRN was involved in AD occurrence. Neuroinflammation plays a critical role in AD (Calsolaro and Edison, 2016). Inflammatory markers of CSF (e.g., transforming growth factor-beta 1 [TGF-β1] and interleukin-10 [IL-10]) or blood (e.g., soluble tumour necrosis factor receptor 1 [sTNFR1] and sTNFR2) were significantly elevated in AD patients compared to healthy controls (Shen et al., 2019). Interestingly, CSF PGRN was also found to be increased over the course of AD (Suarez-Calvet et al., 2018). These lines of evidence suggested a potential link between PGRN and neuroinflammation in AD development, which has not been explored till now. It could be postulated that PGRN might interact with neuroinflammation to contribute to AD pathogenesis, leading to abnormal accumulation of pathological protein, such as β-amyloid (Aβ). To verify this hypothesis, we aimed to (1) examine whether PGRN was associated with inflammatory activities in CNS, (2) explore the roles of neuroinflammation in modulating the influences of PGRN on amyloid pathologies, and (3) investigate the values of PGRN in predict-

List of abbreviations: PGRN, progranulin; CNS, central nervous system; AD, Alzheimer's disease; sTNFR, soluble tumor necrosis factor receptor; ICAM1, intercellular cell adhesion molecule-1; VCAM1, vascular cell adhesion molecule-1; IL, interleukin; CSF, cerebrospinal fluid; Aβ, β-amyloid; ADNI, Alzheimer's Disease Neuroimaging Initiative; CDR, clinical dementia rating; ELISA, enzyme-linked immunosorbent assay; MSD, mass spectrometry detector; CV, coefficient of variation; ECLIA, electrochemiluminescence immunoassays; ADAS, Alzheimer's disease assessment scale; RAVLT, Rey auditory verbal learning test; MMSE, Mini-Mental State Examination; ANCOVAs, one-way analyses of covariance.

* Corresponding author at: Department of Neurology, Qingdao Municipal Hospital, Qingdao University, Qingdao, China, Donghai Middle Road, No.5.
E-mail address: 1037219730@qq.com (W. Xu).

[1] The population data used in preparation for this article were obtained from the Alzheimer's Disease Neuroimaging Initiative (ADNI) database (adni.loni.usc.edu). As such, the investigators within the ADNI contributed to the design and implementation of ADNI and/or provided data but did not participate in the analysis or writing of this report. A complete listing of ADNI investigators can be found at: http://adni.loni.usc.edu/wp-content/uploads/how_to_apply/ADNI_Acknowledgement_List.pdf.

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ing cognitive decline and AD risk, within the framework of A/T/N biomarker profile, using the Alzheimer's Disease Neuroimaging Initiative (ADNI).

2. Methods

2.1. Study participants

ADNI is designed to develop clinical, imaging, genetic, and biochemical biomarkers for the early detection and tracking of AD. The participants are volunteers aged 55-90 years with normal or impaired cognition. Detailed information can be found at http://www.adni-info.org/ and in previous reports (Petersen et al., 2010; Trojanowski et al., 2010; Weiner et al., 2010). At baseline, each participant underwent an in-person interview of general health and functional ability, followed by a standardized assessment including a battery of neuropsychological tests. Follow-up data were collected during evaluations at sequential intervals of approximately 12 months. ADNI was approved by institutional review boards of all participating institutions, and written informed consent was obtained from all participants or their guardians. In the present study, a total of 965 participants who had baseline measurements of CSF PGRN and AD core biomarkers, as well as longitudinal measurements of cognitive functions were included. Among these individuals, 228 had measurements of CSF inflammatory markers.

2.2. Classification methods

The classification methods were in line with 2018 NIA-AA "research framework" for AD diagnosis (Jack et al., 2018). In brief, participants were categorized into specific groups based on biomarker profile as described by the A/T/N scheme (Jack et al., 2016). The A/T/N scheme includes 3 biomarker groups: "A" aggregated amyloid pathology (as indicated by CSF A$\beta_{1\text{-}42}$), "T" aggregated tau (as indicated by CSF p-tau$_{181}$), and "N" neurodegeneration or neuronal injury (as indicated by CSF t-tau). "A+" participants refer to those with CSF A$\beta_{1\text{-}42}$ < 976.6 pg/ml; "T+" those with CSF p-tau$_{181}$ > 21.8 pg/ml; and "N+" those with CSF t-tau > 245 pg/ml. The CSF biomarker statuses established by these cutoffs were proven to be highly concordant with PET classification in ADNI (Hansson et al., 2018). Given that T and N groups were highly correlated, we merged them together to facilitate the analyses, producing a TN group: "TN+" indicates T+ or N+ and "TN-" indicates T- and N- (Suarez-Calvet et al., 2018; Suarez-Calvet et al., 2019).

2.3. CSF measurements of PGRN, inflammatory markers, and AD core biomarkers

CSF procedural protocols in ADNI were described (Shaw et al., 2009). CSF PGRN was measured by a previously reported sandwich immunoassay using the Meso Scale Discovery platform (Capell et al., 2011). All CSF samples were distributed randomly across plates and measured in duplicate. All the antibodies and plates were from a single lot in order to exclude variability between batches. The mean intraplate coefficient of variation (CV) was 2.2%; all duplicate measures had a CV < 15%. PGRN levels were corrected by inter-batch variation and corrected values were used for analyses (for the method see Appendix 1). CSF A$\beta_{1\text{-}42}$, p-tau$_{181}$, and t-tau were analyzed by the electrochemiluminescence immunoassays (ECLIA) Elecsys on a fully automated Elecsys cobas e 601 instrument and a single lot of reagents for each of the three measured biomarkers (provided in UPENNBIOMK9.csv file), as described previously (Hansson et al., 2018). These measurements are for explorative research use only. A total of eight types of CSF inflammatory markers, including 4 anti-inflammatory markers (sTNFR1, sTNFR2, TGF-β1, and IL-10) and four pro-inflammatory markers (intercellular cell adhesion molecule-1 [ICAM1], vascular cell adhesion molecule-1 [VCAM1], IL-6, and IL-7) were measured, using commercially available multiplex immunoassays (Millipore Sigma, Burlington, MA), as described previously (Craig-Schapiro et al., 2011). All samples were run in duplicate along with six standards on each plate. Samples were normalized across plates using CSF standard values. Precision of each analyte was calculated using inter-plate CV < 15%.

2.4. Cognitive measures and AD diagnosis

Global cognitive function was reflected by the total scores of Alzheimer's Disease Assessment Scale (ADAS). Composite scores for executive and memory functions were constructed and validated by referring to the neuropsychological batteries (Crane et al., 2012; Gibbons et al., 2012). Specifically, the indicators of executive functions include Category Fluency, WAIS-R Digit Symbol, Trails A & B, Digit Span Backwards, and clock drawing. The indicators of memory function include relevant items of the Rey Auditory Verbal Learning Test , ADAS, Logical Memory, and Mini-Mental State Examination (MMSE). The Clinical Dementia Rating (CDR) score was used to represent the clinical stage: "0" represents normal cognition, "0.5" represents very mild dementia, and "1" represents mild dementia. The National Institute of Neurological and Communication Disorders/Alzheimer's Disease and Related Disorders Association (NINCDS–ADRDA) criteria (McKhann et al., 1984) was used for the diagnosis of probable AD.

2.5. Statistical analysis

Before the analyses, values as dependent variables were $\log_{10}$-transformed to achieve normal distributions as assessed by Kolmogorov-Smirnov test. All analyses were adjusted for age (continuous variable), gender (female = 1), educational level (continuous variable), *APOE*4 status ("44/34/24" = 1), and CDR score (categorical), except where specifically noted.

First, one-way analyses of covariance (ANCOVAs) were performed to examine the associations of CSF PGRN and CSF inflammatory markers with the A/TN status. Four comparisons were separately conducted for each biomarker group, including A-/TN+ vs. A-/TN-, A+/TN+ vs. A+/TN- (for the associations with tau-related neurodegeneration), A+/TN+ vs. A-/TN+, and A+/TN- vs. A-/TN- (for the associations with amyloid pathology). Next, multiple linear regressions were conducted to explore the associations of PGRN (an independent variable) with neuroinflammatory markers (dependent variables). Furthermore, we explored whether neuroinflammatory markers could modulate the association of PGRN with amyloid pathology. To achieve this, causal mediation analyses were conducted using linear regression models fitted based on the methods proposed by Baron and Kenny (Baron and Kenny, 1986). The direct effects, indirect effects, and the mediating proportion were estimated by Sobel's test (Imai et al., 2010) with the significance determined using 10,000 bootstrapped iterations.

In addition, linear mixed effects (LME) models were used to estimate the longitudinal influences of CSF PGRN on cognitive functions. To facilitate the depiction, CSF PGRN was categorized into three groups (low, moderate, and high) using cutoffs of 1,396 pg/mL and 1,684 pg/mL according to the tertiles of the concentration. The LME models had random intercepts and slopes for time and an unstructured covariance matrix for the random effects, and included the interaction between time (continuous) and the dependent variable (PGRN) as a predictor. Regression diagnostics were conducted and outliers (n = 23) were excluded to indicate that all models met the necessary assumptions: model residuals

Table 1
Demographic and clinical characteristics of the participants.

	Total (n = 228)	A−/TN−(n = 48)	A+/TN−(n = 27)	A+/TN+(n = 120)	A−/TN+(n = 33)
Age, mean (SD), y	74.8 (7.1)	74.5 (6.2)	74.4 (5.8)	74.5 (7.6)	76.5 (7.1)
Female, n (%)	98 (43)	20 (42)	7 (26)	57 (47.5)	14 (42.4)
Education, mean (SD), y	15.5 (3.0)	15.4 (2.8)	15.9 (2.8)	15.4 (3.1)	15.9 (3.1)
*APOE*4 carriers, n (%)	116 (51)	8 (16.7)	15 (56)	87 (72.5)	6 (18.2)
AD diagnosis, n (%)	60 (26.3)	2 (4.2)	7 (26)	46 (38.3)	5 (15.2)
PGRN, mean (SD), pg/ml	1,570 (406)	1,554 (349)	1,521 (330)	1,550 (445)	1,705 (373)
CSF AD core biomarkers, mean (SD), pg/ml					
Aβ42	941 (542)	1,444 (258)	593 (201)	602 (162)	1,725 (556)
P-tau$_{181p}$	29.1 (13.5)	16.9 (2.7)	16.2 (3.7)	36.6 (12.2)	30.1 (12.1)
T-tau	297.5 (116)	192.8 (29)	176.5 (34)	357.5 (102)	331 (106)
CSF neuroinflammatory markers, mean (SD), pg/ml					
sTNFR1	868 (204)	817 (147)	668 (119)	879 (189)	1,067 (199)
sTNFR2	1,035 (251)	949 (180)	804 (130)	1,063 (240)	1,248 (260)
TGF-β1	101.1 (31.6)	96.6 (26.3)	83.0 (26.2)	104 (31.4)	113.7 (36.4)
IL-10	5.65 (2.48)	5.57 (2.53)	5.41 (2.50)	5.65 (2.44)	5.97 (2.62)
ICAM1	374 (192)	327 (127)	331 (156)	379 (198)	461 (248)
VCAM1	41,349 (19,167)	36,343 (11,764)	30,944 (10,796)	40,690 (18,467)	59,522 (23,834)
IL-6	4.5 (2.8)	4.2 (1.8)	3.4 (5.4)	4.5 (2.9)	5.2 (3.6)
IL-7	1.23 (1.00)	1.04 (0.81)	1.13 (0.68)	1.39 (1.15)	1.00 (0.79)

Abbreviations: AD: Alzheimer's disease; SD: standard deviation; PGRN: progranulin; sTNFR: soluble tumor necrosis factor receptor; ICAM1: intercellular cell adhesion molecule-1; VCAM1: vascular cell adhesion molecule-1; IL: interleukin; CSF: cerebrospinal fluid; Aβ: β-amyloid

were normally distributed and did not exhibit heteroscedasticity. Finally, the influence of CSF PGRN on the risk of incident AD was explored using the Kaplan-Meier method. All above analyses were conducted within the framework of A/T/N biomarker profile.

Sensitivity analyses were conducted as follows. (1) the analyses were repeated after excluding outlier values (n = 7) of CSF markers, defined as values situated outside the 3 standard deviations from the mean; (2) rs5848 genotype of *GRN* gene, which was associated with PGRN levels, was added as a covariate in analyses with CSF PGRN as the dependent variable. The results barely changed after these analyses. (3) CDR was considered as a grouping variable for which we found that CDR does not play a significant role when comparing the biomarker levels (**e-Table 1 and e-Fig. 1**).

R version 3.5.1 (packages including "lm", "ggplot2", "mediate", and "nlme") and GraphPad Prism 7.00 software were used for statistical analyses and figure preparation. All tests were two-tailed, with a significance level of $\alpha = 0.05$.

3. Results

3.1. Participant characteristics

A total of 965 participants (44% females, 73.1 ± 7.4 years) were included (e-Table 1), among whom 228 subjects (43% females, 74.8 ± 7.1 years) with neuroinflammation data available were included in the mediation analysis (Table 1). According to the A/TN profile, 48 were categorized within the A-/TN- group, 27 A+/TN-, 120 A+/TN+, and 33 A-/TN+.

3.2. PGRN was associated with neuroinflammatory markers in TN-positive group

We separately draw the distribution patterns of CSF PGRN and 8 marker proteins of neuroinflammation following the A/TN profile. We found CSF levels of 5 out of 8 markers (including sTNFR1, sTNFR2, TGF-β1, ICAM1, and VCAM1) exhibited similar variation tendency with CSF PGRN (Fig. 1). The association analyses indicated that both PGRN and five neuroinflammatory markers were higher in TN+ profile, but lower in A+ group (except for TGF-β1, $p < 0.0001$, e-Table 2), after adjusting for age, gender, education, *APOE*4 status, CDR score, and A/TN status. No significant associations were revealed of A/TN status with IL-6, IL-7, and IL-10 (e-Table 2). We further found that PGRN was positively related to the abovementioned neuroinflammatory markers, but the associations were significant only in TN+ profile (Fig. 2). Interestingly, PGRN showed significant associations with ICAM1 (adjusted $p = 0.03$) and TGF-β1 (adjusted $p = 0.001$) only in A+/TN+ group. These findings strengthened a potentially strong link between PGRN and neuroinflammation in specific populations within the TN+ biomarker profile.

3.3. Neuroinflammation modulated the association of PGRN with lower amyloid burden in TN-positive group

We further asked whether inflammatory markers could modulate the association of PGRN with amyloid pathology. Similarly, positive relationships of CSF Aβ42 with both CSF PGRN (Fig. 3A) and CSF inflammatory markers (Fig. 3B to 3F) were revealed in the TN+, but not in the TN- group. In total population irrespective of the biomarker framework, the mediation analyses indicated that the association of PGRN with alleviated cerebral amyloid deposition was modulated by specific neuroinflammatory markers, including sTNFR1 (proportion = 50.3%, $p = 0.006$), sTNFR2 (proportion = 28.4%, $p = 0.01$), and VCAM1 (proportion = 44.2%, $p = 0.008$) (e-Figure 2). These results remained significant after Bonferroni correction. Within the biomarker framework, the abovementioned mediation effects of neuroinflammation (sTNFR1, sTNFR2, and VCAM1) were significant only in TN+ profile, with the mediation proportion ranged from 30% to 60% (e-Fig. 3). Similar results were obtained in A+/TN+ group (Fig. 3G): ICAM1 and TGF-β1 were specifically revealed as mediating molecules for the association between PGRN and amyloid burden in A+/TN+ group (e-Fig. 4).

3.4. CSF PGRN predicted slower cognitive decline and lower risk of AD in TN-positive group

Based on the above findings, it could be postulated that the roles of PGRN in protecting AD or cognitive decline might be, at least partially, influenced by the TN status. To verified this hypothesis, the following analyses were conducted. We explored whether the values of CSF PGRN for predicting longitudinal changes of cog-

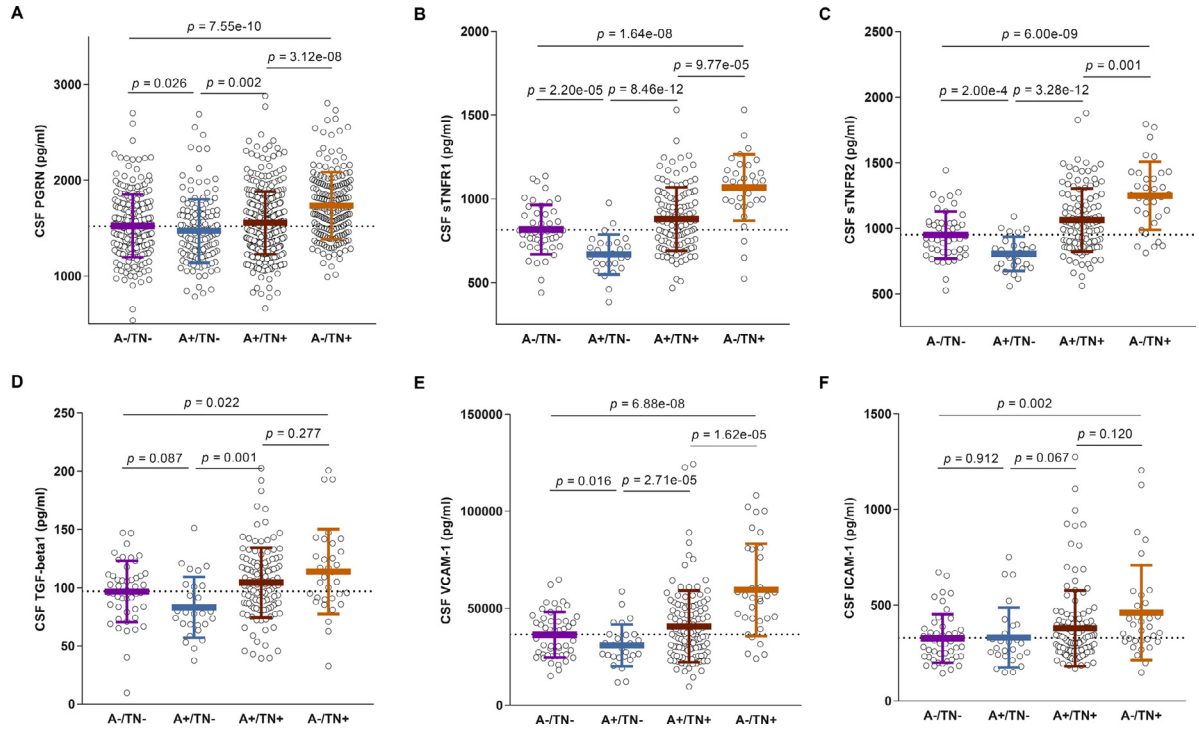

Fig. 1. Early increases of CSF PGRN and CSF five neuroinflammatory markers were associated with tau-related neurodegeneration.
Scatter plot depicting CSF levels of PGRN and five neuroinflammatory markers (including sTNFR1, sTNFR2, TGF-β1, ICAM1, and VCAM1) for each of the four biomarker profiles, as defined by the A/T/N framework. The T (tau pathology) and N (neurodegeneration) group were merged because these two biomarker groups were highly correlated. Solid bars represent the mean and the standard deviation (SD). P-values were assessed by one-way ANCOVAs adjusted for age, gender, educational level, and CDR score.
Abbreviations: A: Aβ pathology biomarker status; T: tau pathology biomarker status AD; N: neurodegeneration biomarker status; Alzheimer's disease; CDR: clinical dementia rating; CSF: cerebrospinal fluid.

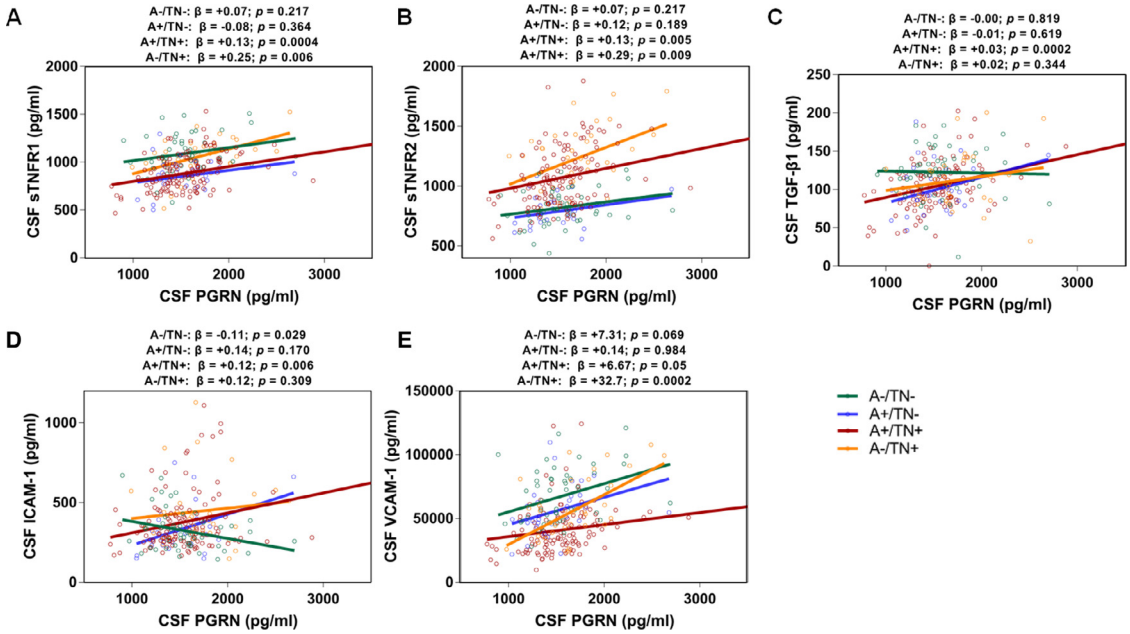

Fig. 2. Relationship of CSF PGRN with CSF neuroinflammatory markers.
The x axis represents CSF PGRN level and the y axis represents CSF specific neuroinflammatory marker level. Each A/T/N biomarker profile is represented in a different color: A-/TN- are depicted in green, A+/TN- in blue, A+/TN+ in dark red, and A-/TN+ in orange. P-values were assessed by multiple linear regression models adjusted for age, gender, educational level, and CDR score.

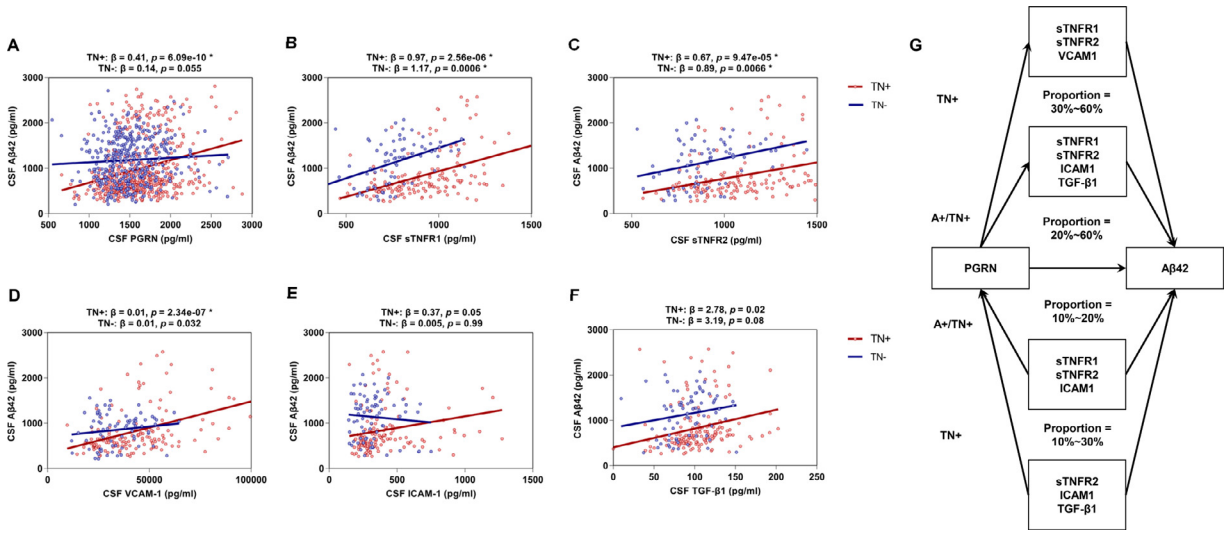

Fig. 3. Neuroinflammation modulates the association of PGRN with ameliorated cerebral amyloid-β burden in TN+ population
The association of CSF PGRN (3A) and abovementioned CSF neuroinflammatory markers (3B to 3F) with CSF Aβ42 were only significant or stronger in TN+ group compared to TN- group. The mediating effects of neuroinflammatory markers (including sTNFR1, sTNFR2, and VCAM1) on the relationships of CSF PGRN with CSF Aβ42 were significant only in TN+ profile, with the mediation proportion ranged from 30% to 60% (3G). Interestingly, we also found smaller (10%~30%) but significant mediation effects of CSF PGRN in influencing association of CSF neuroinflammatory markers (sTNFR1, sTNFR2, and ICAM1) with CSF Aβ42 in TN+ profile (3G). Similar results were obtained in A+/TN+ group (3G). P-values were adjusted for age, gender, education, *APOE*4 status, and CDR score. * the results survived the Bonferroni correction.

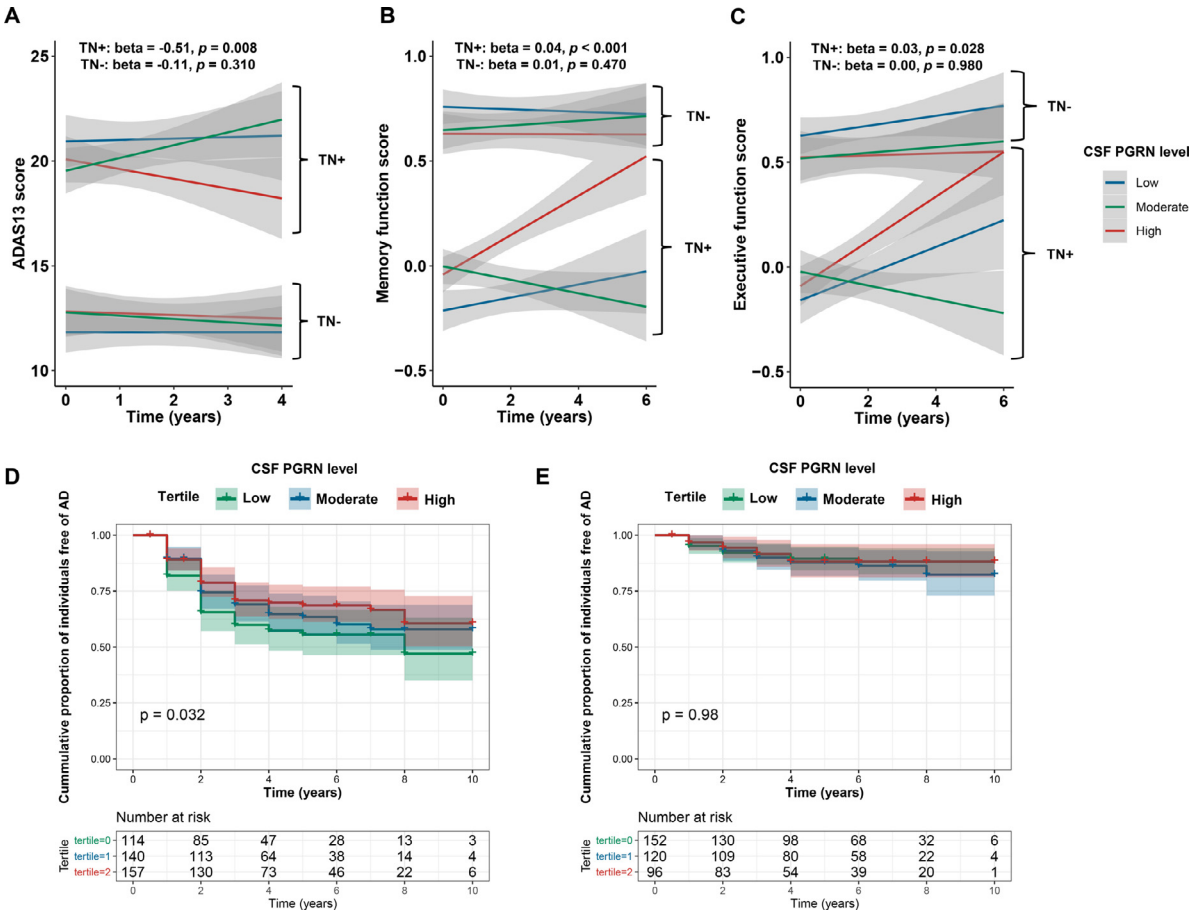

Fig. 4. Values of CSF PGRN in predicting cognitive decline and incident risk of AD, stratified by the TN status
CSF PGRN were categorized into three tertiles (low, moderate, and high level) in order to facilitate the drawing. *p*-values were assessed by mixed-effect models adjusted for age, gender, education, *APOE*4 status, CDR, and amyloid status (A profile). We found protective roles of high CSF levels of PGRN in preventing decline of cognitive functions, including the general cognition (A), memory function (B), and executive function (C), in TN+ but no TN- profile. Moreover, higher CSF PGRN was associated with lower risk of incident AD in TN+ profile (D), but not in TN- profile (E).

nitive functions were influenced by the TN status. We found protective roles of CSF PGRN in cognitive function, including the general cognition ($p = 0.008$, Fig. 4A), memory function ($p = 0.0002$, Fig. 4B), and executive function ($p = 0.028$, Fig. 4C) only in TN+ group. Furthermore, higher CSF PGRN was associated with lower risk of incident AD in TN+ group (Fig. 4D), but not in TN- group (Fig. 4E), as revealed by the ADNI cohort of 779 nondemented samples who were followed up to 10 years.

4. Discussion

Herein, we were the first to explore the relationships of PGRN with neuroinflammatory makers in CNS and evaluate their synergetic mediating effects on amyloid pathology. Our results indicated that neuroinflammation might modulate the association of PGRN with amyloid pathologies and the mediating associations were limited to TN+ group. The predicting values of PGRN for cognitive decline or AD were also constrained to individuals who are suffering from neurodegeneration due to neuronal damages (TN-positive group).

PGRN was proposed to be a hallmark of microglia-mediated neuroinflammation (Suarez-Calvet et al., 2018). Similar to PGRN, CSF sTREM2, a marker of microglial activation, was found to be elevated in early AD with TN+ profile (Suarez-Calvet et al., 2019). It was reported that CSF PGRN was associated with CSF sTREM2 only in AD and non-Alzheimer's disease pathophysiology groups (Suarez-Calvet et al., 2018), suggesting PGRN might be a hallmark of neuroinflammation occurring with neurodegeneration. Though no causal conclusion can be made due to the cross-sectional design, these findings indicated a close relationship between PGRN and neuroinflammation in neurodegeneration.

Neuroinflammation plays a critical role in modulating AD pathologies. We and other teams previously reported increased peripheral levels of sTNFR1, sTNFR2 (Buchhave et al., 2010; Shen et al., 2019; Zhang et al., 2014) and IL-6, as well as elevated CSF levels of IL-10 and TGF-β1 in AD compared with the controls (Shen et al., 2019). TNFRs could be activated by binding of soluble TNF, a hallmark of neuroinflammation as well as neurodegenerative conditions (McCoy and Tansey, 2008), and could be cleaved to generate sTNFRs. The circulating levels of sTNFR were positively associated with the levels of plasma amyloid and tau (Buchhave et al., 2010; Zhang et al., 2014) and the conversion rate to dementia (Buchhave et al., 2010). Another study on transgenic mice showed TNFR1 deletion reduced Aß pathology, microglia activation, neuron loss, and memory deficits (He et al., 2007). In concordance with the present study, previous studies found CSF levels of ICAM1 and VCAM1 were increased during the preclinical and prodromal stages of AD (Janelidze et al., 2018; Rauchmann et al., 2020).

It was found that PGRN could suppress neuroinflammation following induced toxic stimuli or injury (Ma et al., 2017; Martens et al., 2012). Our results suggested PGRN might interact with specific neuroinflammatory markers to reduce amyloid burden. This suggests that inflammation activities might play a "double-edged sword" role in dealing with neurodegeneration. Similar clues were reported for microglia, which adopted numerous fates with homeostatic microglia and a microglial neurodegenerative phenotype representing two opposite ends (Götzl et al., 2019). Another possible explanation is that increased PGRN could be a counter response to the elevated inflammatory markers to counteract their detrimental consequences. More experiments are needed to validate these assumptions.

We found PGRN and specific neuroinflammatory markers were higher in individuals with TN+ profile and lower in those with A+ profile. The reasons may be that (1) an increase of CSF PGRN can be a direct consequence of microglial expression or a consequence of neuronal cell death releasing PGRN into neuropil, and that (2) PGRN and specific neuroinflammatory markers were involved in the metabolism of amyloid pathology, such as the clearance of aggregated amyloid via normal immune activation and lysosomal functioning. Future *in vitro* studies are needed to verify these clinical findings. Moreover, our results indicated that the values of higher levels of PGRN in predicting lower AD risk were constrained to those who had significant neuronal damages, which needed to be verified in future larger studies. However, it is still unclear whether regulating PGRN in TN+ status could confer benefits to lower amyloid burden and AD risk. Aβ plaques with PGRN were identified in low-plaque individuals, suggesting PGRN was involved in early plaque formation. However, the impacts of PGRN on AD pathologies, especially β-amyloid (Aβ), was disputable in in-vivo or in-vitro studies. Takahashi et al., reported PGRN deficiency significantly reduces diffuse amyloid plaque growth (Takahashi et al., 2017), and Hosokawa et al., also reported PGRN haploinsufficiency reduced amyloid beta deposition in APP mouse model (Hosokawa et al., 2018). On the contrary, Minami et al., (Minami et al., 2014) and Van Kampen JM et al., (Van Kampen and Kay, 2017) reported overexpression of PGRN reduced amyloid burden. Considering predicting values of PGRN varied according to the TN status, future experiments might need to consider this condition for development of precision medicine of AD.

Additionally, it is noteworthy that PGRN is primarily a marker of lysosomal functioning (Paushter et al., 2018) besides neuroinflammation. Disturbances of lysosomal function can result in multiple pathological features, such as disordered clearance and abnormal accumulations of insoluble proteins (e.g., amyloid and tau), increased autophagy, etc (Colacurcio et al., 2018). Therefore, future studies are warranted to explore whether PGRN could influence AD via such pathways as neuronal survival, autophagy, and blood brain barrier (BBB) integrity.

5. Limitation

Caution is warranted given that several limitations existed for the present study. The mediation associations only reflect but cannot equal to the causal relationships. Longitudinal cohort as well as well-designed experiments should be conducted to verify our findings about the influences of PGRN on neuroinflammatory markers and AD pathologies. No experiments were conducted in the present study. The in vivo and in vitro studies are thus warranted to examine the causal relationships of neuronal injuries or tau-related neurodegeneration with PGRN, neuroinflammatory markers, and amyloid metabolism.

6. Conclusions

Our study provided preliminary clues linking PGRN to neuroinflammatory activities in TN+ populations. PGRN could interact with neuroinflammation to influence amyloid burden. The relationships were restricted to those with neurodegenerative changes and might help lower risks of cognitive decline and AD. However, the causal relationship warrant verification in future experiments.

Verification

1. There are no actual or potential conflicts of interest for all authors or their institutions.
2. No sources of financial support related to the manuscript being submitted.

3. The data contained in the manuscript being submitted have not been previously published, have not been submitted elsewhere and will not be submitted elsewhere while under consideration at Neurobiology of Aging.
4. ADNI was approved by institutional review boards of all participating institutions, and written informed consent was obtained from all participants or their guardians.
5. All authors have reviewed the contents of the manuscript being submitted, approve of its contents and validate the accuracy of the data.

Ethics approval and consent to participate

ADNI was approved by institutional review boards of all participating institutions, and written informed consent was obtained from all participants or their guardians.

Consent for publication

Not applicable

Availability of data and materials

The datasets used and/or analyzed during the current study are available from http://adni.loni.usc.edu/

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Authors' contributions

Dr. Wei Xu: conceptualization and design of the study, collection and analysis of the data, drafting and revision of the manuscript, and prepared all the figures. Dr. Chen-Chen Tan, MS. Xi-Peng Cao, and Prof. Lan Tan: revision of the manuscript.

Competing interests

The authors declare that they have no competing interests.

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Supplementary materials

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