Aging and Disease
www.aginganddisease.org
Early access date: May 15, 2026
http://dx.doi.org/10.14336/AD.2025.1587

Original Article

Deep Venous Perivascular Space Dysfunction Reflects Glymphatic Aging and Predicts Cognitive Vulnerability: *In Vivo* Human Evidence

Kemeng Zhang[1#], Ying Zhou[1#], Yifei Li[1], Xin Huang[1], Mengmeng Fang[2], Kai Wei[2], Min Lou[1*]

[1]Department of Neurology, Second Affiliated Hospital of Zhejiang University, School of Medicine, State Key Laboratory of Transvascular Implantation Devices, Hangzhou, China. [2]Department of Radiology, Second Affiliated Hospital of Zhejiang University, School of Medicine, Hangzhou, China

[Received December 30, 2025; Revised April 16, 2026; Accepted April 17, 2026]

ABSTRACT: Impaired glymphatic clearance has been recognized as a fundamental mechanism of cognitive decline. While models suggest cerebrospinal fluid influx along arterial perivascular spaces (aPVS) and efflux along venous PVS (vPVS), their differential roles in humans are unclear. We aimed to visualize directional glymphatic flow in vivo, classify human PVS functionally and determine their associations with cognition. This study included 91 patients undergoing intrathecal gadodiamide with serial MRI at baseline, 4.5 hours, 15 hours and 39 hours post-injection. PVS showing early (4.5h) and delayed (39h) enhancement peaks were defined as aPVS and vPVS, respectively. Among 742 basal ganglia (BG) and 1380 centrum semiovale (CSO) PVS analyzed, 10.4% and 21.6% were aPVS, while 62.7% and 52.0% were vPVS, respectively. BG-vPVS burden correlated with age (r=0.275, p<0.001) and hypertension. Among 60 patients with cognitive assessment (telephone Montreal Cognitive Assessment, T-MoCA) data, only BG-vPVS burden independently correlated with lower scores after adjusting for age and education (β=-0.16, p=0.041). This study provided direct in vivo MRI evidence of glymphatic flow within human PVS. We introduced a novel functional classification method to differentiate arterial from venous PVS, finding their different role in cognitive impairment, which may represent a potential target for therapeutic intervention.

Keywords: perivascular spaces, glymphatic flow, cognition

INTRODUCTION

Cognitive impairment and dementia, including Alzheimer disease (AD), constitute major contributors to disability and mortality worldwide. Beyond amyloid-β (Aβ) deposition and tau aggregation[1], emerging evidence underscores the critical role of impaired metabolic waste clearance in the progression of cognitive decline. The recently identified glymphatic system, which facilitates cerebrospinal fluid (CSF) and interstitial fluid exchange, plays a central role in clearing neurotoxic solutes such as Aβ and tau from the brain parenchyma.

In rodent models, glymphatic transport follows a directional circuit in which CSF enters via arterial perivascular spaces (aPVS) and exits through venous perivascular spaces (vPVS) toward meningeal lymphatic drainage[2, 3]. Age-related vascular stiffening, hypertension, and the loss of astroglial aquaporin-4 (AQP4) polarization are known to disrupt this flow, leading to solute stagnation and neuroinflammation[2]. Despite the established conceptual framework in animals, direct evidence of functional arterial and venous separation in human glymphatic transport and its relevance to cognition remains limited.

Human neuroimaging studies have primarily focused on the static morphological burden of PVS, which correlates with aging and small vessel disease[4, 5]. However, such structural markers cannot distinguish

***Correspondence should be addressed to:** Dr. Min Lou, Second Affiliated Hospital of Zhejiang University, School of Medicine, Hangzhou, China. Email: lm99@zju.edu.cn; loumingxc@vip.sina.com. #These authors contributed equally to this work.

Copyright: © 2026 Zhang K. et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

between impaired inflow and outflow, nor do they reflect dynamic solute transport. Identifying which component of the glymphatic pathway—arterial influx or venous efflux—is preferentially compromised may reveal early mechanisms of clearance dysfunction and provide new therapeutic opportunities for cognitive preservation.

In this study, we employed serial three-dimensional T1-weighted magnetic resonance imaging (3D-T1 MRI) following intrathecal gadolinium administration to dynamically visualize glymphatic tracer movement in vivo. By quantifying enhancement timing, we established a functional classification of human PVS as either arterial or venous, thereby bridging structural imaging with physiological understanding. We further examined how these distinct PVS subtypes relate to cognitive performance across vascular risk profiles.

METHODS

Participants

This prospective observational study enrolled patients aged over 18 years old who underwent lumbar puncture for clinical indications and voluntarily participated in the study between April 2018 and May 2024. Exclusion criteria included known hypersensitivity to contrast agents, a history of severe allergic reactions, renal dysfunction, pregnancy or lactation, and absence of visible PVS in the basal ganglia or centrum semiovale on baseline MRI.

All enrolled patients underwent T2 weighted imaging (T2WI) and T2 Fluid attenuated inversion recovery (T2-FLAIR) at baseline, and 3D-T1 at baseline and multiple time points (4.5 hours, 15 hours, and 39 hours) after intrathecal administration of an MRI contrast agent (gadodiamide). The 4.5-hour and 39-hour imaging time points were selected a priori based on preclinical and human glymphatic imaging studies demonstrating early CSF influx and delayed clearance phases following intrathecal tracer administration. However, we acknowledge that the true peak signal intensity may occur between the sampled time points, which may introduce uncertainty in peak identification and affect classification accuracy. All patients were admitted and observed in the ward for at least 39 hours after intrathecal injection. Demographic and vascular risk factors were recorded, including age, sex, years of education, hypertension, diabetes mellitus, smoking, and alcohol consumption.

MRI protocol

All scans were performed using a 3.0T MRI scanner (GE 750, GE Healthcare, Chicago, IL) with identical acquisition parameters across time points. The imaging parameters were as follows: Sagittal head 3D-T1: repetition time = 7.3 ms, echo time = 3.0 ms, flip angle = 8°, thickness = 1 mm, field of view = 25 × 25 cm^2, matrix = 250 × 250. Axial T2: repetition time = 2500 ms, echo time = 4 ms/100 ms, thickness = 4 mm, field of view = 24 × 24 cm^2, matrix = 512 × 512. Axial T2-FLAIR: repetition time = 8400 ms, echo time = 150 ms, field of view = 24× 24 cm^2, matrix = 256 × 256, inversion time = 2100 ms, slice thickness = 4 mm, and in-plane resolution = 0.47 × 0.47 mm.

Intrathecal Administration of Gadodiamide

Intrathecal gadodiamide was administered under clinical indication and ethically monitored; no serious adverse event occurred. Intrathecal injection was performed at the L3-4 or L4-5 intervertebral space using a standard lumbar puncture technique. After confirming correct needle placement by observing CSF backflow, 1mL of 0.5mmol/mL gadodiamide[6] (Omniscan; GE Healthcare) was slowly injected. Following needle withdrawal, patients were asked to gently rotate their bodies twice along the longitudinal axis and then remain supine for at least 4 hours after the procedure.

Image Analysis

Evaluation of MRI-visible PVS

PVS in the basal ganglia (BG-PVS) and centrum semiovale (CSO-PVS) were evaluated on baseline T2WI. For each region, the slice showing the highest PVS burden was selected for analysis. A neurologist (Z.K.) with 10 years of experience, blind to all clinical data, graded the number of PVS on the most affected side using a validated 5-point scale[7]: Grade 0, no PVS can be found; Grade 1, 1-10 PVS; Grade 2, 11-20 PVS; Grade 3, 21-40 PVS; Grade 4, >40 PVS.

Evaluation of Arterial and Venous PVS

All T2WI and 3D T1 images were co-registered using SPM12 (Wellcome Department of Neurology, University College of London, UK). Given the known asymmetry of MRI-visible perivascular spaces, analyses were performed in the hemisphere with the highest PVS burden to capture the region of maximal glymphatic dysfunction and to avoid dilution of associations arising from bilateral averaging. Regions of interest (ROI) were manually placed on unilateral BG-PVS and CSO-PVS at the slice with maximal burden. Mean signal intensity of each ROI was measured at all time points, and signal unit ratios (SURs) were normalized to the sagittal sinus to reduce inter-scan variability[8]. However, because the sagittal

sinus itself may exhibit delayed contrast accumulation following intrathecal administration, normalization using this reference may introduce shared temporal variation with PVS signal intensity. Therefore, SUR and PC values should be interpreted as relative signal measures rather than absolute indices of glymphatic influx or clearance. Percentage change (PC) in SUR from baseline to each post-contrast time point was calculated, and the time of maximum PC was defined as the peak enhancement time. PVS peaking at 4.5 hours were classified as arterial PVS (aPVS), whereas those peaking at 39 hours were defined as venous PVS (vPVS). Analyses were performed separately for BG and CSO regions. Perivascular spaces demonstrating peak enhancement at the intermediate 15-hour time point were considered to have ambiguous enhancement kinetics and were therefore excluded from classification. While this approach was intended to reduce misclassification, it may also introduce selection bias by excluding a substantial proportion of PVS with intermediate or mixed enhancement profiles.

Cognitive Assessment

We assessed the cognitive function using telephone version of the Montreal Cognitive Assessment (T-MoCA) with total score of 22 points[9]. This version includes attention, calculation, language, abstraction, recall, and orientation items, omitting visuospatial and executive function subtests (e.g., cube drawing, trail making test, clock drawing) that require visual or written responses. Total and subdomain scores were recorded for each participant.

Statistical Analysis

Statistical analyses were performed on SPSS software version 22.0 (IBM Corporation). All data were subject to Kolmogorov-Smirnov tests for normality. Correlations were determined by Kendall's Tau correlation coefficient. Differences between groups were determined using rank sum tests or Chi-Square test. Linear regression analysis was used to assess independent associations between cognitive function and PVS burden, adjusting for covariates with $P < 0.05$ in univariate analysis. All statistical tests were two-tailed, with a significance level of 0.05.

Ethical Approval

Written informed consent was obtained from all participants prior to enrollment. The study protocol, including intrathecal gadolinium administration, was approved by the Human Ethics Committee of the Second Affiliated Hospital of Zhejiang University. All clinical investigation has been conducted according to the principles expressed in the Declaration of Helsinki.

RESULTS

Patients' Characteristics

A total of 91 patients (44 [48.4%] female, age was 57.9 ± 1.3 years) were included. The final diagnosis included peripheral neuropathy (n = 49), neurodegenerative diseases (n = 17), encephalitis (n = 15), normal pressure hydrocephalus (n = 4), suspected CSF leakage (n = 5), hepatic encephalopathy (n = 1). The prevalence of hypertension, diabetes mellitus, smoking, and alcohol use were present in 47.3%, 34.1%, 35.2%, and 26.4% of patients, respectively.

The average number of BG-PVS and CSO-PVS was 8.2 ± 1.7 and 15.2 ± 0.8, respectively. Three (3.3%), 70 (76.9%), 10 (11.0%), 7 (7.7%) and 1 (1.1%) patients presented BG-PVS as grade 0, 1, 2, 3 and 4, while 31 (34.1%), 31 (34.1%) and 29 (31.9%) patients had CSO-PVS as grade 1, 2, and 3, respectively. No patient presented CSO-PVS as grade 0 and 4. Three patients without visible BG-PVS were excluded from BG flow analysis.

Dynamic Signal Changes in PVS

After intrathecal contrast administration, signal intensity within both BG-PVS and CSO-PVS increased over time (Fig. 1). Within the same individual, distinct PVS often exhibited variable enhancement profiles, indicating heterogeneous flow dynamics among different PVS (Fig. 1). The mean SUR in both BG-PVS and CSO-PVS progressively increased from baseline to 15 hours and further to 39 hours ($P < 0.001$ for both comparisons, Fig. 2).

Functional Classification of Arterial and Venous PVS

Among 742 individual BG-PVS, 77 (10.4%) reached peak enhancement at 4.5 hours and were classified as arterial-type PVS (BG-aPVS), while 465 (62.7%) peaked at 39 hours, denoting venous-type PVS (BG-vPVS). Similarly, among 1,380 CSO-PVS, 298 (21.6%) were CSO-aPVS and 717 (52.0%) CSO-vPVS. PVS peaking at 15 hours accounted for 26.9% of BG-PVS and 26.4% of CSO-PVS and were excluded from subsequent aPVS/vPVS analyses. These findings indicate that the majority of visible PVS, particularly in the basal ganglia, exhibit delayed enhancement consistent with a venous drainage profile.

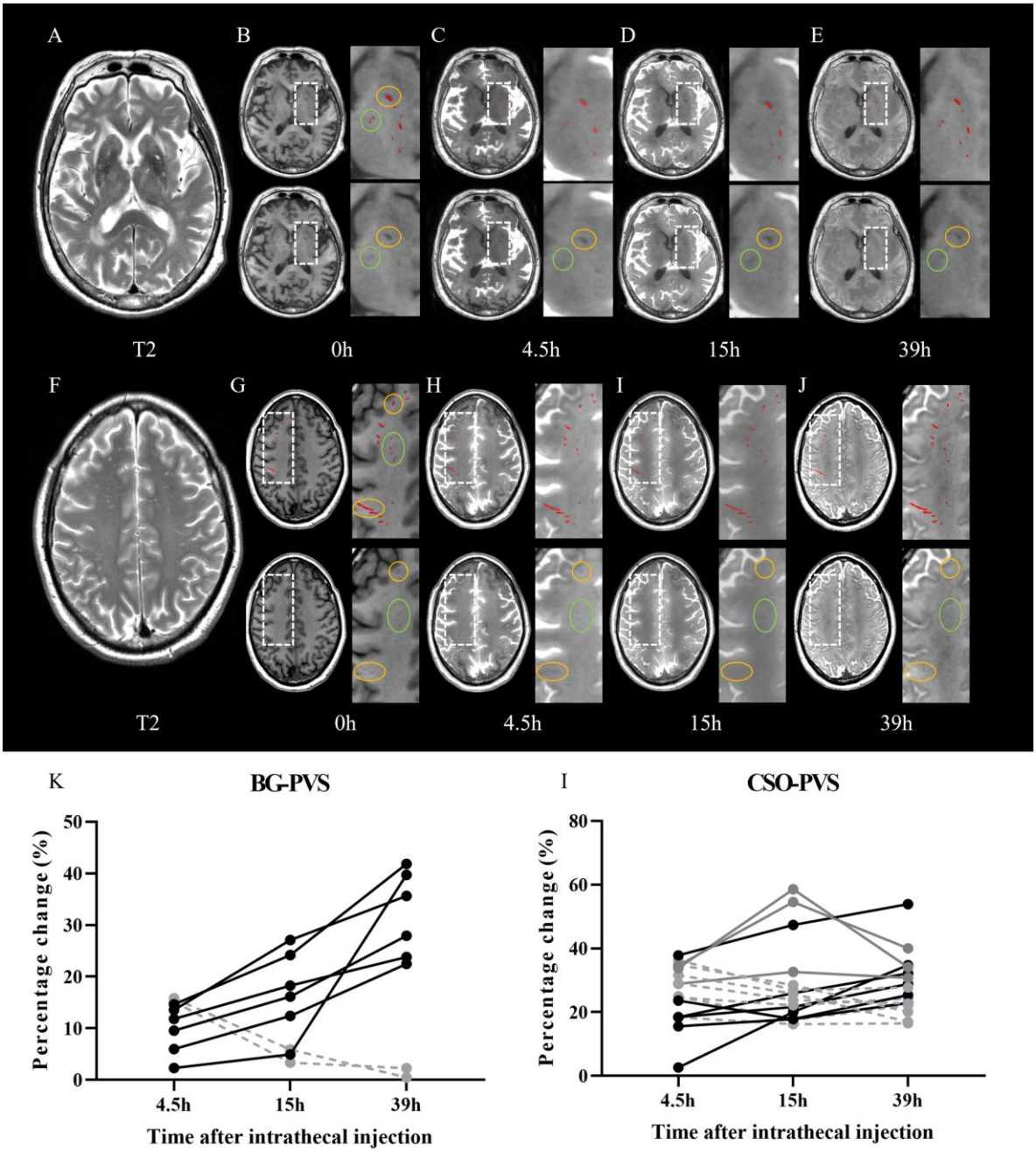


Figure 1. Representative images showing signal intensity change at different time points in MRI-visible perivascular spaces in basal ganglia (BG-PVS) and centrum semiovale (CSO-PVS). The upper image shows a patient with BG-PVS of grade 1 on axial T2-image **(A)**. Glymphatic function was evaluated on T1-image before **(B)**, and 4.5 h **(C)**, 15 h **(D)** and 39 h **(E)** after intrathecal injection. The regions of interest of BG-PVS are marked in red color (upper) with enlarged view (right column). Significant signal intensity change of BG-PVS could be observed at different time points after intrathecal injection. The single PVS (yellow circle) shows the highest signal intensity at 39 hours, while other PVS (green circle) shows the highest at 4.5 hours. The lower image shows a patient with CSO-PVS of grade 2 on axial T2-image **(F)**. Glymphatic function was also evaluated on T1-image **(G-J)**. Significant signal intensity change of CSO-PVS could be observed at different time points after intrathecal injection, marked in red color (upper) with enlarged view (right column). Some PVS (yellow circles) show the highest signal intensity at 39 hours, while other PVS (green circles) shows the highest at 4.5 hours. The percentage change (PC) of the signal unit ratio (SUR) in each BG-PVS **(K)** and CSO-PVS **(I)** on the above representative images from baseline to different time points after intrathecal injection shows that the time point at which the PC of the SUR in each PVS was highest could be at 4.5h (dotted gray line), 15h (solid gray line) or 39h (solid dark line).

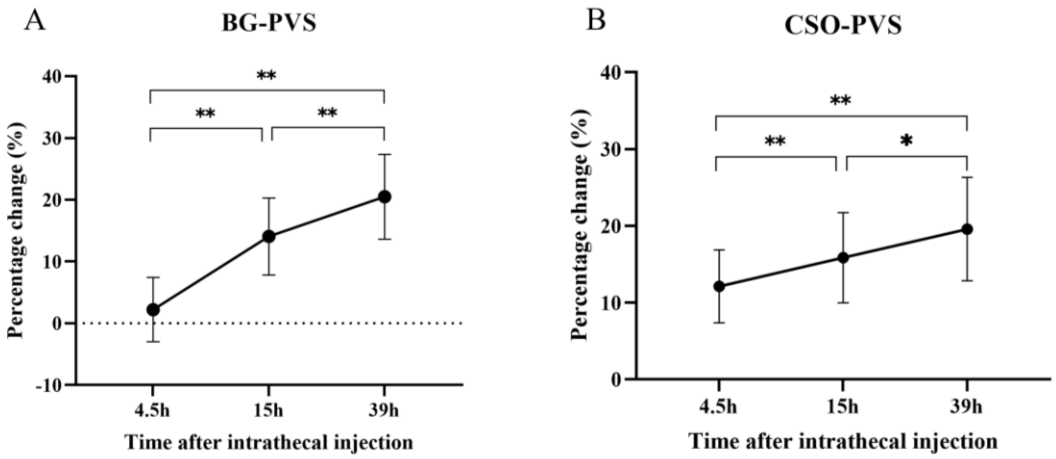


Figure 2. The percentage change (PC) of the signal unit ratio (SUR) in MRI-visible perivascular spaces in basal ganglia (BG-PVS) and centrum semiovale (CSO-PVS) from baseline to different time points after intrathecal injection in the entire cohort of 91 individuals. The PC of the SUR in BG-PVS **(A)** and CSO-PVS **(B)** was increased at 15 and 39 hours compared with that at 4.5 hours after intrathecal injection in the entire study cohort (n = 91) (paired Wilcoxon signed rank test; *$P < 0.01$, **$P < 0.001$). Error bars refer to 95% confidence intervals (CI).

Factors Associated with PVS Subtypes

Among the four PVS subtypes, only BG-vPVS showed a significant positive correlation with age (r = 0.275, $P < 0.001$). BG-aPVS (r = -0.025, $P = 0.764$), CSO-aPVS (r = 0.010, $P = 0.703$), and CSO-vPVS (r = 0.060, $P = 0.417$) were not significantly associated with age (Figure 3). Patients with a history of hypertension exhibited significantly higher burdens of BG-vPVS (median [IQR], 6.0 [2.5-8.5] vs 1.0 [0.0-5.25], $P < 0.01$) and CSO-vPVS (7.0 [2.5-14.0] vs 3.0 [1.0-7.25], $P = 0.025$) compared with those without hypertension. No significant differences were found for aPVS burden across hypertension groups (all $P > 0.05$; Fig. 4; Supplemental Table 1).

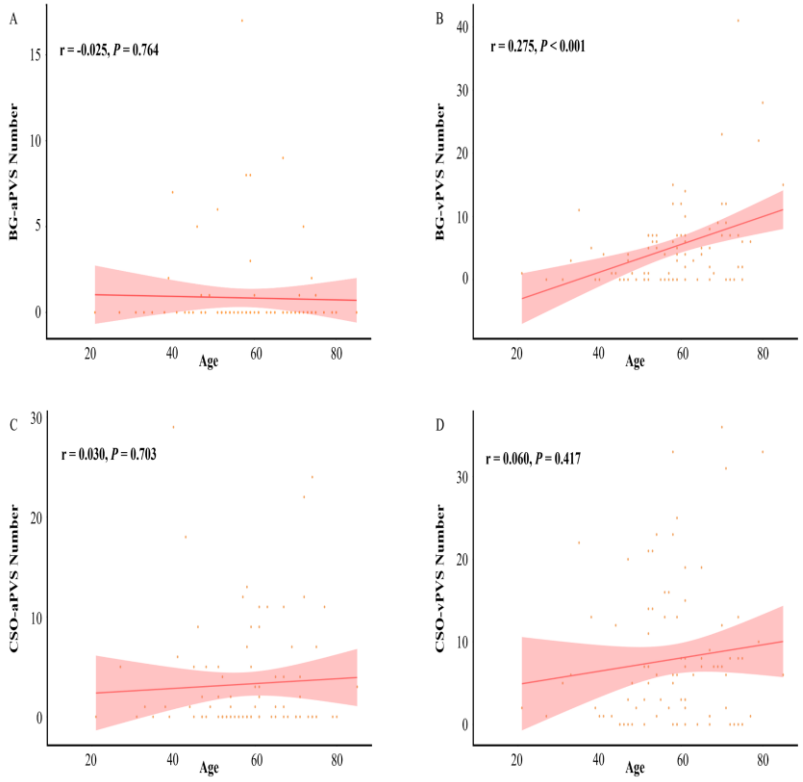


Figure 3. Correlation analysis of age and MRI-visible perivascular spaces in basal ganglia and centrum semiovale. Among the four PVS subtypes, only BG-vPVS **(B)** showed a significant positive correlation with age (Kendall's Tau correlation coefficient, r = 0.275, $P < 0.001$) in the entire study cohort (n=91). BG-aPVS (Kendall's Tau correlation coefficient, r = -0.025, $P = 0.764$), CSO-aPVS (Kendall's Tau correlation coefficient, r = 0.010, $P = 0.703$), and CSO-vPVS (Kendall's Tau correlation coefficient, r = 0.060, $P = 0.417$) were not significantly associated with age **(A, C, D).** BG-aPVS, arterial perivascular spaces in the basal ganglia; BG-vPVS, venous perivascular spaces in the basal ganglia; CSO-aPVS, arterial perivascular spaces in the centrum semiovale; CSO-vPVS, venous perivascular spaces in the centrum semiovale.

Other vascular risk factors, including diabetes mellitus, smoking, and alcohol use, were not significantly related to PVS subtype burden (all $P > 0.05$; Supplemental Table 1).

Relationship between PVS Subtypes and Cognitive Function

Finally, 60 patients completed the T-MoCA test, while among the other 31 patients, 12 refused to undergo the test and 19 were lost to follow-up. No significant differences were found in age, sex, hypertension prevalence, diabetes status, or BG/CSO-PVS burden (all $P > 0.05$) between the T-MoCA completed (n=60) and non-completed (n=31) subgroups (Supplemental Table 2). Among those who completed the T-MoCA, only BG-vPVS burden was significantly associated with lower cognitive scores (r = -0.244, $P = 0.010$; Fig. 5; Supplemental Table 3). This association remained significant after adjusting for age and years of education (β = -0.16, $P = 0.041$; Figure 5). However, this association did not consistently remain significant after correction for multiple comparisons. No significant correlations were observed between cognitive scores and the burden of BG-aPVS, CSO-aPVS, or CSO-vPVS (all $P > 0.05$).

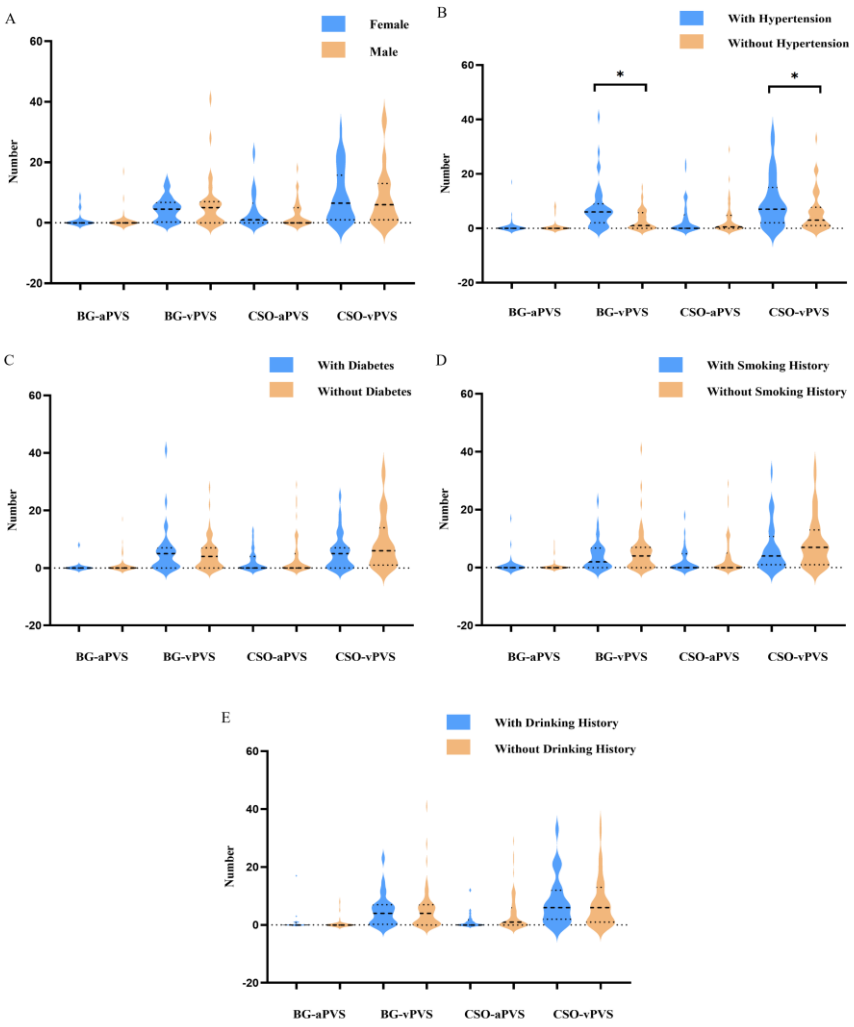


Figure 4. Relationship between patients' characteristics and MRI-visible perivascular spaces in basal ganglia and centrum semiovale. In the entire study cohort (n=91), patients with a history of hypertension **(B)** exhibited significantly higher burdens of BG-vPVS (median [IQR], 6.0 [2.5-8.5] vs 1.0 [0.0-5.25], rank sum test, $P < 0.01$) and CSO-vPVS (7.0 [2.5-14.0] vs 3.0 [1.0-7.25], rank sum test, $P = 0.025$) compared with those without hypertension. No significant differences were found for aPVS burden across hypertension groups **(A, C-E)**. Other vascular risk factors, including diabetes mellitus, smoking, and alcohol use, were not significantly related to PVS subtype burden (rank sum test, all $P > 0.05$). BG-aPVS, arterial perivascular spaces in the basal ganglia; BG-vPVS, venous perivascular spaces in the basal ganglia; CSO-aPVS, arterial perivascular spaces in the centrum semiovale; CSO-vPVS, venous perivascular spaces in the centrum semiovale. *P < 0.01. Error bars refer to 95% confidence intervals (CI).

DISCUSSION

In this prospective study utilizing serial intrathecal contrast-enhanced MRI, we demonstrated that temporal enhancement patterns can differentiate arterial from venous PVS in the human brain. Among the four PVS subtypes evaluated, only BG-vPVS showed significant associations with both aging and cognitive decline. These findings not only provide in vivo support for a directional glymphatic flow in humans but also indicate that dysfunction of venous efflux, rather than arterial influx, is likely a critical determinant of impaired brain clearance during aging and vascular disease.

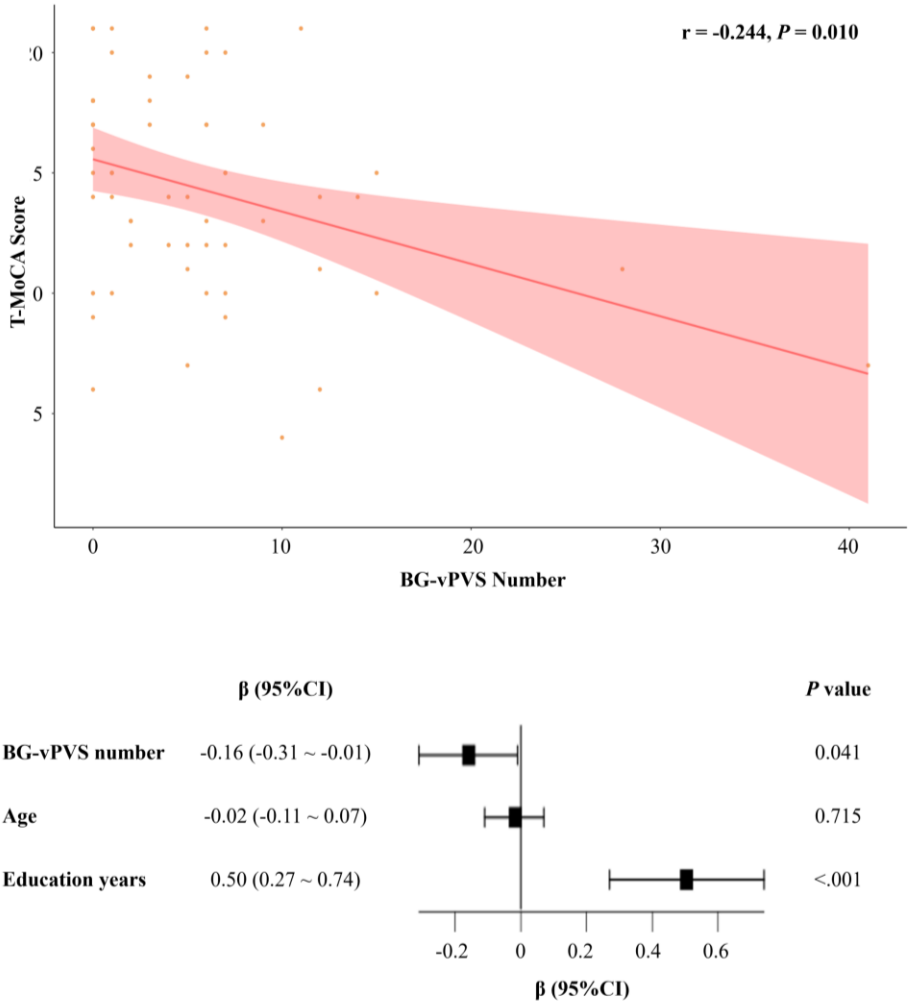


Figure 5. Analysis of the relationship between T-MoCA score and venous MRI-visible perivascular spaces in basal ganglia. Among the 60 patients who completed the T-MoCA, only BG-vPVS number was significantly correlated with T-MoCA score (Kendall's Tau correlation coefficient, r = -0.244, $P = 0.010$). After adjusting for age and education years, BG-vPVS number was still related with T-MoCA score (linear regression, β = -0.16, $P = 0.041$). T-MoCA, telephone version for the Montreal Cognitive Assessment; BG-vPVS, venous perivascular spaces in the basal ganglia.

Animal experiments have shown that glymphatic flow follows a defined path, entering along arterial perivascular spaces and exiting through venous ones[2]. Our dynamic MRI results extend this concept to humans, demonstrating distinct enhancement peaks consistent with temporal CSF influx and interstitial efflux. Although gadolinium-based tracers may remain detectable for extended periods, the stepwise increase in signal from 4.5 to 39 hours confirms active solute transit rather than passive stagnation, supporting the feasibility of this imaging-based classification and providing a practical framework for investigating glymphatic dynamics *in vivo*.The predominance of venous-type PVS observed in our cohort may reflect the central role of venous outflow in maintaining interstitial homeostasis. This venous predominance could represent either a compensatory enlargement in response to increased drainage demand or, alternatively, a vulnerability of the venous outflow pathway to age-related structural changes. Age-related collagen deposition, reduced vascular pulsatility, and AQP4 depolarization can slow interstitial drainage along perivenous routes, leading to metabolic by-product accumulation, glial activation, and perivascular congestion. These processes may contribute to what can be described as a "glymphatic aging" phenotype, characterized by a progressive decline in glymphatic function associated with vascular and cellular alterations(e.g., astroglial AQP4 depolarization, cerebral vascular stiffening, reduced vascular pulsatility). However, given the cross-sectional design and cohort heterogeneity, this term should be interpreted as a conceptual framework rather than direct mechanistic evidence. In this process, venous efflux failure acts as an upstream biological hallmark preceding structural neurodegeneration. Moreover, the basal ganglia may be particularly vulnerable due to its vascular anatomy and metabolic demand. This region is supplied by deep perforating arteries and drained by small medullary veins with limited collateralization. Hypertension and small vessel disease further narrow perivenous spaces or reduce their compliance, disproportionately impeding clearance in the basal ganglia. In contrast, the centrum semiovale drains mainly to cortical veins, where compliance and pulsatility may remain relatively preserved. These findings align with diffusion tensor MRI studies reporting reduced perivascular diffusivity and slower solute kinetics in small vessel disease[10]. It should be noted that direct in vivo measures of astroglial AQP4 polarization, cerebral vascular stiffening, or other molecular biomarkers of glymphatic function were not available in the present clinical study, due to ethical constraints and the lack of validated non-invasive imaging tools in humans. Therefore, the associations observed in this study should not be interpreted as direct mechanistic evidence of glymphatic aging. Rather, BG-vPVS burden may be regarded as a surrogate in vivo imaging phenotype reflecting impaired glymphatic efflux in humans, which is consistent with, but does not directly demonstrate, underlying molecular or cellular mechanisms.

Prior human studies have mainly described PVS morphology rather than function. Conventional MRI often captures tubular, vessel-aligned aPVS more prominently than vPVS, contributing to earlier assumptions that most visible PVS are arterial. However, contrast-tracking methods such as ours assess physiological transit rather than static appearance, revealing that functionally active PVS are predominantly venous. This observation challenges the traditional anatomical interpretation and underscores the need to differentiate structural visibility from dynamic functionality in future PVS research.

The observed link between BG-vPVS burden and cognitive performance suggests a potential of venous PVS as an imaging biomarker for early cognitive vulnerability. This association persisted after adjusting for age and education, suggesting a physiological rather than purely demographic mechanism. Monitoring venous PVS burden may therefore help identify individuals at risk for cognitive decline before the onset of overt dementia. Therapeutically, interventions aimed at improving glymphatic efflux, such as optimizing sleep, enhancing vascular pulsatility, or restoring AQP4 polarity, may hold greater promise than those targeting arterial inflow alone. Future longitudinal studies are needed to determine whether venous PVS dysfunction predicts progression to mild cognitive impairment or dementia, and whether its recovery can mitigate cognitive deterioration.

Several limitations warrant consideration. First, this study included patients with various diseases. The heterogeneity of the diseases as well as the individual variations may have influenced glymphatic flow dynamics. Occult blood-brain-barrier dysfunction or focal inflammation could either amplify or mask true glymphatic differences. Although consistent associations between BG-vPVS burden, aging, hypertension, and cognition were observed, these findings should be interpreted with caution given the heterogeneity of the study population. In sensitivity analyses, these associations were attenuated and did not consistently retain statistical significance in more homogeneous subsets, suggesting that the observed relationships may reflect a composite influence of both age-related and disease-related alterations rather than a purely aging-driven glymphatic process. The reproducibility of these associations suggests that the observed glymphatic signature is not merely disease-specific but rather represents a fundamental aging-linked clearance phenotype. Future prospective studies in community-

based aging cohorts are warranted to validate temporality and prognostic value. Second, the limited temporal sampling (4.5 h, 15 h, and 39 h) represents an inherent constraint of the current study. Because the true peak enhancement of individual PVS may occur between these intervals, the classification into aPVS and vPVS may be subject to misclassification. Moreover, the aPVS/vPVS classification should be interpreted as an operational imaging-based construct reflecting temporal enhancement patterns, rather than a definitive representation of arterial or venous physiology. Future studies with higher temporal resolution are warranted to better capture the full dynamics of glymphatic tracer movement and to validate the robustness of this classification approach. Third, the use of the sagittal sinus as a reference region may introduce shared temporal variance with perivascular signal changes, representing a methodological limitation of the present study. Although alternative reference regions such as normal-appearing white matter may be considered, no consensus reference standard currently exists for intrathecal glymphatic MRI in humans. Fourth, we analyzed the hemisphere with the highest PVS burden, which does not account for bilateral or contralateral involvement and may introduce potential selection bias. Future studies incorporating bilateral or regional PVS metrics are warranted to further validate the observed associations. Fifth, T-MoCA, although validated by telephone, lacked visuospatial and executive subtests and may underestimate mild impairment. The modest but significant association between BG-vPVS burden and T-MoCA scores (β=-0.16) is likely an underestimate of the true association between BG-vPVS dysfunction and cognitive decline, as the T-MoCA does not assess the basal ganglia's key cognitive roles. Future studies should employ comprehensive in-person cognitive assessments (including validated visuospatial and executive function subtests) to fully characterize the relationship between BG-vPVS dysfunction and basal ganglia-related cognitive decline. Sixth, the exclusive use of gadodiamide necessitates caution, as different contrast agents might yield divergent results.

In summary, this study provided direct in vivo MRI evidence of glymphatic flow within human PVS. We introduced a novel functional classification method to differentiate arterial from venous PVS, finding their different role in cognitive impairment, which may represent a potential imaging marker for further investigation into therapeutic strategies targeting glymphatic function.

Acknowledgements

This study was supported by the National Natural Science Foundation of China (82571465, U23A20426) and Research Project on Prevention and Treatment Techniques for Stroke of Development Center for Medical Science & Technology, National Health Commission of the People's Republic of China (WKZX2023CZ0220).

Author Contribution statement

Z.K. drafted the manuscript, participated in study design and data collection, conducted the statistical analyses. Z.Y. drafted the manuscript, participated in study design and data collection. L.Y. and H.X. participated in data collection. F.M., and W.K. participated in designing the MRI sequences and data collection. L.M. participated in study design and data collection, data interpretation and made a major contribution in revising the manuscript.

Conflicts of Interest

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Supplementary Materials

The Supplementary data can be found online at: www.aginganddisease.org/EN/10.14336/AD.2025.1587.

References

[1] Ross CA, Poirier MA (2004). Protein aggregation and neurodegenerative disease. Nat Med, 10 Suppl:S10-17.

[2] Jessen NA, Munk AS, Lundgaard I, Nedergaard M (2015). The Glymphatic System: A Beginner's Guide. Neurochem Res, 40(12):2583-2599.

[3] Hablitz LM, Pla V, Giannetto M, Vinitsky HS, Staeger FF, Metcalfe T, et al. (2020). Circadian control of brain glymphatic and lymphatic fluid flow. Nat Commun 11(1):4411.

[4] Wardlaw JM, Benveniste H, Nedergaard M, Zlokovic BV, Mestre H, Lee H, et al. (2020). Perivascular spaces in the brain: anatomy, physiology and pathology. Nat Rev Neurol, 16(3):137-153.

[5] Deike-Hofmann K, Reuter J, Haase R, Paech D, Gnirs R, Bickelhaupt S, et al. (2019). Glymphatic Pathway of Gadolinium-Based Contrast Agents Through the Brain: Overlooked and Misinterpreted. Invest Radiol, 54(4):229-237.

[6] Zhou Y, Cai J, Zhang W, Gong X, Yan S, Zhang K, et al. (2020). Impairment of the Glymphatic Pathway and Putative Meningeal Lymphatic Vessels in the Aging Human. Ann Neurol, 87(3):357-369.

[7] Potter GM, Chappell FM, Morris Z, Wardlaw JM (2015). Cerebral perivascular spaces visible on magnetic resonance imaging: development of a qualitative rating

scale and its observer reliability. Cerebrovasc Dis, 39(3-4):224-231.

[8] Eide PK, Vatnehol SAS, Emblem KE, Ringstad G (2018). Magnetic resonance imaging provides evidence of glymphatic drainage from human brain to cervical lymph nodes. Sci Rep, 8(1):7194.

[9] Pendlebury ST, Welch SJ, Cuthbertson FC, Mariz J, Mehta Z, Rothwell PM (2013). Telephone assessment of cognition after transient ischemic attack and stroke: modified telephone interview of cognitive status and telephone Montreal Cognitive Assessment versus face-to-face Montreal Cognitive Assessment and neuropsychological battery. Stroke, 44(1):227-229.

[10] Chen X, Wang J, Shan Y, Cai W, Liu S, Hu M, et al. (2019). Cerebral small vessel disease: neuroimaging markers and clinical implication. J Neurol, 266(10):2347-2362.