




# Age-Related Association Between Circulating Inflammatory Indicators and Plaque Enhancement on High-Resolution Magnetic Resonance Imaging in Patients with Intracranial Atherosclerotic Stenosis

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**Background:** Plaque enhancement is a non-specific marker of local inflammatory response, which may offer additional insights together with circulating inflammatory markers. Few studies have analyzed the association between intracranial atherosclerotic stenosis (ICAS) plaque enhancement and circulating inflammatory markers. Given the age-related variability in the progression of ICAS, this study aims to explore the association between the two across different age groups.

**Methods:** This retrospective study recruited 120 patients with ICAS-related ischemic events who had undergone high-resolution magnetic resonance imaging. Plaque enhancement index at the most stenosed site of the culprit vessel was calculated. Levels of circulating inflammatory indicators, including high-sensitivity C-reactive protein (hsCRP), lymphocyte-to-white blood cell ratio (LWR), systemic immune inflammation index (SII), platelet-to-lymphocyte ratio (PLR), lymphocyte-to-monocyte ratio (LMR), and neutrophil-to-lymphocyte ratio (NLR), were detected. General linear regression models were established to analyze the association between ICAS plaque enhancement index and circulating inflammatory indicators.

**Results:** In this study, hsCRP, but not other circulating inflammatory indicators, had a significant positive association with ICAS plaque enhancement index ( $\beta=0.219$ , 95% CI [0.036, 0.349],  $P=0.02$ ). After multivariate adjustment, there was still a marginal correlation between hsCRP and the enhancement index ( $\beta=0.220$ , 95% CI [0.025, 0.362],  $P=0.05$ ). The association was particularly significant in patients <60 years rather than those  $\geq 60$  years. For participants <60 years, hsCRP had the highest contribution to plaque enhancement interpretation.

**Conclusion:** ICAS plaque enhancement index was positively associated with hsCRP, particularly in participants aged <60 years. This may be helpful for understanding the significance of the enhancement index in clinical practice.

**Keywords:** circulating inflammatory indicators, high-sensitivity C-reactive protein, plaque enhancement index, intracranial atherosclerosis

## Introduction

Stroke is the second leading cause of death and third leading cause of disability worldwide. Ischemic stroke accounts for the majority of strokes and is prone to severe neurological impairment.<sup>1,2</sup> Intracranial atherosclerotic stenosis (ICAS) is

an important cause of ischemic stroke.<sup>3,4</sup> Ischemic stroke caused by atherosclerotic diseases is not only associated with the degree of stenosis, but also closely related to the composition of plaque.<sup>5-7</sup> Then came the concept of vulnerable plaque. Plaque enhancement identified by high-resolution magnetic resonance imaging (HR-MRI) is considered a major characteristic of vulnerability plaque.<sup>8</sup> ICAS plaque enhancement was an independent risk factor for acute ischemic stroke, and can predict stroke recurrence.<sup>9-11</sup> However, its clinical application is largely limited by its non-specificity.

Previous studies on coronary and carotid plaques have shown that plaque enhancement on HR-MRI is indicative of local inflammation.<sup>9,12</sup> There is substantial evidence supporting the association between circulating inflammation and the local inflammatory response in atherosclerotic plaques.<sup>13,14</sup> The combination of circulating inflammation indicators and plaque enhancement can better characterize plaque instability thus has better clinical applications.<sup>15,16</sup> This has sparked interest in the relationship between circulating inflammatory markers and plaque enhancement. Several indicators can quantitatively assess the level of systemic inflammatory response, including traditional markers such as high-sensitivity C-reactive protein and novel indicators represented by the ratios of blood cells, such as the neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR), lymphocyte-to-monocyte ratio (LMR), and the systemic immune inflammation index (SII). However, the relationship between circulating inflammatory indicators and local plaque inflammation in ICAS remains poorly investigated. The progression of intracranial arteries significantly differs from those of extracranial arteries, such as the carotid and coronary arteries. Typically, ICAS progresses slowly during middle age, with a noticeable increase in severity after the age of sixty.<sup>17</sup> Therefore, in this study, we explored the relationship between ICAS plaque enhancement index obtained by HR-MRI and circulating inflammatory indicators in different age groups (<60 years vs  $\geq$ 60 years), trying to provide help for better understanding the significance of ICAS plaque enhancement.

## Methods

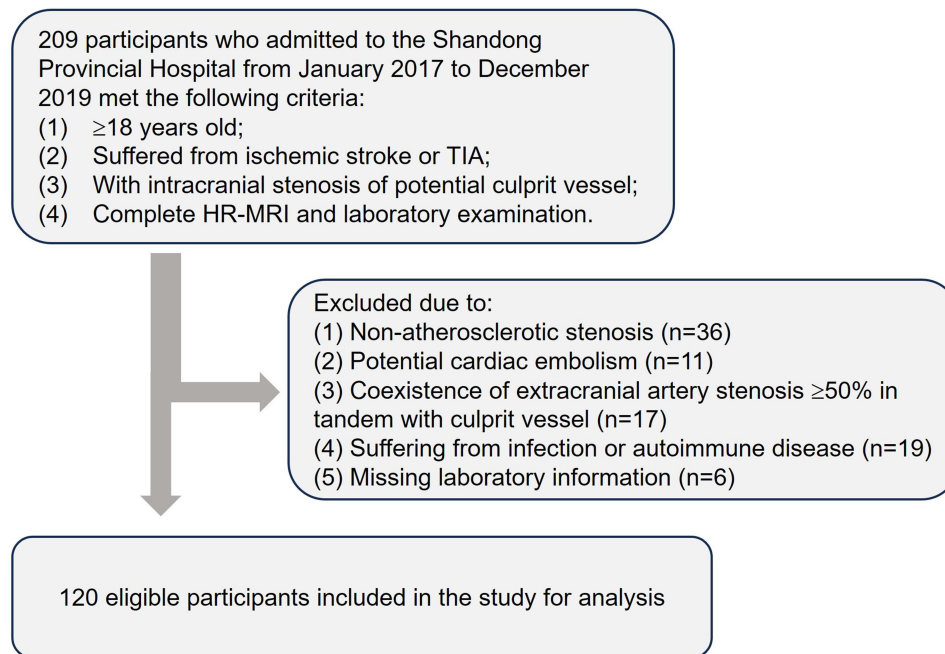
### Study Population

This retrospective study included patients admitted to Shandong Provincial Hospital, Shandong University, between January 2017 and December 2021, for research purposes. The inclusion criteria were as follows: (1)  $\geq$ 18 years of age; (2) had suffered from ischemic stroke or TIA; (3) intracranial stenosis (including intracranial segment of the internal carotid artery, intracranial segment of the vertebral artery, middle cerebral artery, and basilar artery) of the culprit vessel confirmed by MRA, CTA, or DSA; and (4) complete HR-MRI and laboratory examination. The exclusion criteria were as follows: (1) non-atherosclerotic vascular lesions, such as arterial dissection, vasospasm, vasculitis, arterial aneurysm, or moyamoya disease; (2) potential cardiac embolism; (3)  $\geq$ 50% extracranial stenosis in tandem with the culprit vessel; (4) acute or chronic infection or autoimmune diseases; and (5) unqualified HR-MRI data or missing other essential information. Among the 209 patients, 36 were excluded because of non-atherosclerotic stenosis, 11 due to potential cardiac embolism, 17 due to extracranial stenosis ( $\geq$ 50%), 19 due to infection or autoimmune diseases, and 6 due to incomplete data for analysis, resulting in the remaining 120 patients for analysis (Figure 1). This study was approved by the Ethics Committee of Shandong Provincial Hospital, Shandong University. All the participants signed an informed consent form for the study.

### High-Resolution Vessel Wall MR Imaging

HR-MRI was collected within 48h of the patient's arrival at the hospital and was performed using a 3.0 T MRI scanner (Achieve; Philips Medical Systems, Best, The Netherlands) with a sensitivity encoding (SENSE) parallel imaging head coil. The sequences included transverse 3D T1-weighted imaging (T1WI) and Volumetric ISotropically Turbo spin-echo acquisition (VISTA) sequences both before and after contrast administration.<sup>11</sup>

The T1 VISTA scanning parameters were as follows: time of repetition (TR)/time of echo (TE), 800 ms/18 ms; echo column length (ETL), 16; field of view (FOV), 200 mm  $\times$  180 mm  $\times$  40 mm; layer thickness (Thk), 0.3 mm; matrix, 332 $\times$ 302 (isotropic spatial resolution: 0.3  $\times$  0.3 $\times$ 0.3 mm); average, 1-2; parallel imaging (SENSE) factor, 2 (along the phase encoding direction); and scanning time, 378s. Sensitized blood flow compensation was used to suppress the intraluminal blood signals, and a 90° refocus flip angle was used to enhance the air flow effect and to reduce images.



**Figure 1** The flow chart of the study.

**Abbreviations:** TIA, transient ischemic attack; HR-MRI, high-resolution magnetic resonance imaging.

Gadopentetate dimeglumine (Magnevist; Schering, Berlin, Germany) was administered intravenously (0.1 mmol per kilogram of body weight), and T1 VISTA enhancement sequences was repeated 5 minutes after contrast administration. The scan parameters and sites were consistent with T1 VISTA sequence.

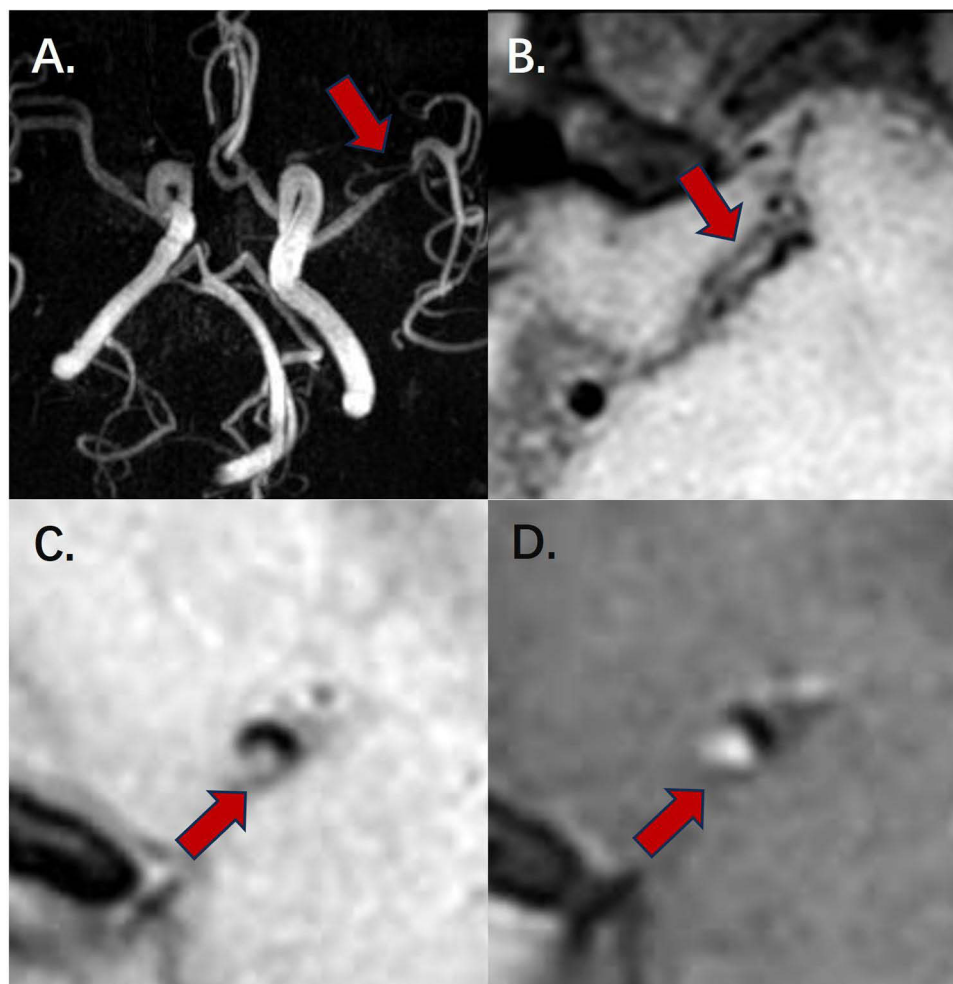
MR vessel wall imaging was independently evaluated by two experienced neuroradiologists using Medical Imaging Viewer software (Extended MR WorkSpace, Philips Medical Systems). A plaque was considered a culprit plaque when it was (a) the only lesion within the vascular territory of the stroke or (b) the most stenotic lesion when multiple plaques were present within the same vascular territory of the stroke.<sup>18</sup> The narrowest site of atherosclerotic lesion was inferred according to the largest lumen stenosis on T1WI. **Figure 2** shows high-resolution magnetic resonance imaging of a plaque in the left middle cerebral artery. The intraclass correlation coefficient (ICC) values were 0.951 (95% CI 0.858–0.980) for stenosis degree, 0.939 (95% CI 0.821–0.975) for plaque burden, 0.952 (95% CI 0.881–0.979) for plaque enhancement index, and 0.933 (95% CI 0.866–0.967) for the remodeling index. The mean values of measurements from two individuals were used for the analysis in this study.

Plaque enhancement was quantified according to the signal intensity of the plaque ( $SI_{\text{plaque}}$ ) and gray matter ( $SI_{\text{gray matter}}$ ) on pre- and matched post-contrast T1WI using the following formula.<sup>19,20</sup>

The vessel-cerebrospinal fluid interface was used to manually track the vascular area (VA), and the blood-intima interface was used to determine the lumen area (LA). The vessel wall area (WA) was calculated as the VA minus the LA. Plaque burden was calculated as the ratio of plaque area and WA.<sup>20</sup> The remodeling index was the ratio of the VA to that of the  $VA_{\text{reference}}$  (VA of the non-diseased part of the proximal portion of the narrowest site).<sup>21</sup> The degree of stenosis was calculated as  $(1 - LA/LA_{\text{reference}})$  (LA of the non-diseased part of the proximal portion of the narrowest site)  $\times 100\%$ .<sup>21</sup> According to the degree of stenosis, participants were further divided into mild stenosis ( $< 50\%$ ) and moderate-to-severe stenosis ( $\geq 50\%$ ).

## Assessment of Circulating Serum Inflammatory Indicators

Fasting venous blood samples were collected from patients within 48 h of HR-MRI examination. All serological tests were conducted in the laboratory department of Shandong Provincial Hospital, Shandong University. The serum hsCRP concentration was determined by immunoturbidimetry, with a detectable concentration  $> 0.01$  mg/L. Total white blood



**Figure 2** High-resolution magnetic resonance imaging of a plaque in the left middle cerebral artery.

**Notes:** (A) Overview of Magnetic Resonance Angiography. The red arrow indicates the narrowest site of the culprit vessel. (B) Overview of the stenosis site at left middle cerebral artery in the pre-contrast T1 axial section. The red arrow indicates the stenosis site. (C) Imaging of the narrowest site at left middle cerebral artery in the pre-contrast T1 sagittal section. The red arrow indicates the culprit plaque at the stenosis site. (D) Imaging of the narrowest site at left middle cerebral artery in the post-contrast T1 sagittal section. The red arrow indicates the culprit plaque at the stenosis site.

cells, neutrophils, lymphocytes, monocytes, and platelets were obtained from the blood cell counts. Lymphocyte-to-white blood cell ratio (LWR) was calculated using the lymphocyte-to-white blood cell ratio. The SII was calculated as the platelet count  $\times$  neutrophil/lymphocyte count. PLR was calculated as the ratio of platelets to lymphocytes, LMR as the ratio of lymphocytes to monocytes, and NLR as the ratio of neutrophils to lymphocyte.

## Covariates Assessment

Clinical data were recorded, including age, sex, smoking habit, drinking habit, hypertension, diabetes mellitus (DM), low-density lipoprotein cholesterol (LDL-c), and time from the onset of stroke symptoms to HR-MRI collection. Smoking habit was defined as current smoking within 6 months. Drinking habit was defined as alcohol consumption within six months. Hypertension was defined as a systolic blood pressure  $\geq 140$  mmHg and/or diastolic blood pressure  $\geq 90$  mmHg, use of antihypertensive medication, or a self-reported history of hypertension. DM was defined as fasting plasma glucose (FPG)  $\geq 7.0$  mmol/L, use of any type of hypoglycemic medication, or self-reported history of DM.

## Statistical Analysis

The characteristics of the participants are expressed as mean and standard deviation (SD) values for continuous variables with normal distributions, median and interquartile range (IQRs) for continuous variables with non-normal distribution,

and frequencies and percentages for categorical variables. ANOVA and post-hoc pairwise comparisons (LSD) (for continuous variables with a normal distribution) or the Mann–Whitney *U*-test (for continuous variables with a non-normal distribution) were used to compare the characteristics between mild stenosis and moderate-to-severe stenosis. The chi-square test or Fisher's exact test was used for categorical variables. The inter-observer agreement of continuous variables was evaluated by ICC using a two-way random model with absolute agreement. A linear regression model was employed to examine the association between circulating inflammatory indicators and the plaque enhancement index. Continuous variables with skewed distributions were logarithmically converted for the analysis. Age, sex, hypertension, DM, smoking habit, drinking habit, LDL-c level, time interval between HR-MRI and stroke, and degree of stenosis were defined as covariates for adjustment. The regression model was expressed with a  $\beta$  coefficient and a 95% confidence interval (95% CI). The contributions of different indicators in the model were calculated as a percentage of the sum of the squared coefficients. SPSS (version 26.0, IBM Corp., Armonk, NY, USA) was used for analysis, and a two-tailed *P* value <0.05, was considered statistically significant.

## Results

### Baseline Characteristics of Study Participants

This study included 120 eligible participants, of which 70 (58.5%) were male. The average age of the study population was 61.5 (IQR 52.3–68.8) years, with 52 participants <60 years and 68 participants  $\geq$ 60 years. Circulating inflammation indicators and plaque characteristics on HR-MRI are shown in Table 1. For participants aged <60 years and  $\geq$ 60 years, there were no significant differences between the two age groups in clinical characteristics such as sex, hypertension, DM, smoking habit, and drinking habit (Table 1). Similarly, there were no significant differences in plaque characteristics, including stenosis degree, plaque burden, plaque enhancement index, time interval between HR-MRI and stroke, and circulating inflammation indicators, such as hsCRP, LWR, SII, PLR, LMR, and NLR (Table 1).

**Table 1** Characteristics of the Study Participants

Characteristics	Total (n=120)	<60 years (n=52)	$\geq$ 60 years (n=68)	<i>P</i> value
Age, years	61.5 (52.3–68.8)	51.0 (46.0–54.0)	67.5 (63.0–72.0)	–
Male, n (%)	70 (58.3%)	31 (59.6%)	39 (57.4%)	0.80
Smoking habit, n (%)	44 (36.7%)	21 (40.4%)	23 (33.8%)	0.46
Drinking habit, n (%)	46 (38.3%)	24 (46.2%)	22 (32.4%)	0.12
Hypertension, n (%)	82 (68.3%)	33 (63.5%)	49 (72.1%)	0.32
DM, n (%)	31 (25.8%)	13 (25.0%)	18 (26.5%)	0.86
LDL-c, mmol/L	2.43 (1.97–2.96)	2.50 (2.04–3.04)	2.37 (1.95–2.82)	0.27
hsCRP, mg/L	1.26 (0.67–2.83)	1.21 (0.62–2.67)	1.51 (0.73–3.53)	0.37
LWR	0.28 (0.23–0.33)	0.28 (0.23–0.33)	0.28 (0.23–0.33)	0.55
SII	474.45 (370.22–722.43)	461.14 (354.89–784.38)	498.40 (374.81–707.34)	0.62
PLR	125.10 (100.71–160.43)	118.96 (93.88–156.51)	128.69 (108.67–161.01)	0.12
LMR	4.20 (3.09–5.08)	4.20 (3.43–5.18)	4.22 (2.98–5.08)	0.46
NLR	2.21 (1.65–2.87)	2.18 (1.57–2.82)	2.27 (1.73–2.91)	0.46
Stenosis degree	67.29 (50.58–78.91)	67.72 (52.19–80.86)	65.97 (48.77–77.75)	0.50
Stenosis degree				0.81
Mild (<50%)	29 (24.2%)	12 (23.1%)	17 (25.0%)	
Moderate-to-severe ( $\geq$ 50%)	91 (75.8%)	40 (76.9%)	51 (75.0%)	
Plaque burden	71.07 (55.88–85.65)	67.83 (57.25–86.42)	73.24 (53.22–84.86)	0.64
Remodeling index	1.00 (0.80–1.20)	0.98 (0.72–1.22)	1.02 (0.82–1.18)	0.72
Enhancement index	57.11 (32.67–99.76)	61.19 (39.96–101.70)	47.59 (25.96–99.76)	0.29
Time interval between HR-MRI and stroke, days	21.5 (13.0–47.0)	21.5 (12.0–59.8)	21.5 (14.0–45.5)	0.80

**Note:** Data was shown as numbers (%) for category variables, median (first-third quartiles) for continuous variables.

**Abbreviations:** DM, diabetes mellitus; LDL-c, Low-density lipoprotein cholesterol; hsCRP, high-sensitivity C-reactive protein; LWR, lymphocyte-to-white blood cell ratio; SII, systemic immune inflammation index; PLR, platelet-to-lymphocyte ratio; LMR, lymphocyte-to-monocyte ratio; NLR, neutrophil-to-lymphocyte ratio.



## Association Between Plaque Enhancement Index and Circulating Inflammation Indicators

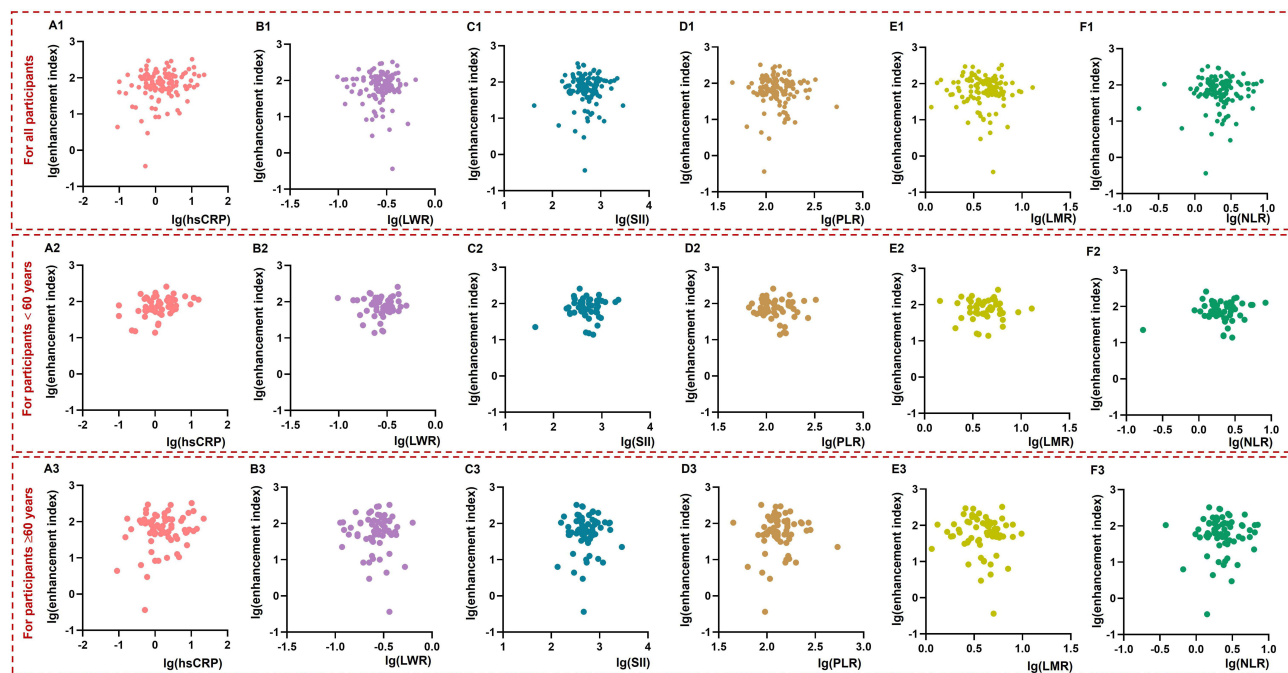
The association between circulating inflammatory indicators and the plaque enhancement index is shown in Figure 3. Specifically, hsCRP was positively associated with plaque enhancement index ( $\beta=0.219$ , 95% CI [0.036, 0.349],  $P=0.02$ ). However, no significant associations were found between plaque enhancement index and other circulating inflammatory indicators, such as LWR, SII, PLR, LMR, and NLR (Table 2). Even after adjusting for age, sex, hypertension, DM, low-density lipoprotein cholesterol, smoking habit and drinking habit, hsCRP was still independently associated with the plaque enhancement index ( $\beta=0.222$ , 95% CI [0.025, 0.364],  $P=0.03$ ). Additionally, a marginally significant association was observed after adjusting for the stenosis degree and time interval between HR-MRI and stroke ( $\beta=0.190$ , 95% CI [-0.002, 0.337],  $P=0.05$ ) (Table 2).

Notably, the association between hsCRP and the plaque enhancement index was only significant for patients <60 years ( $\beta=0.381$ , 95% CI [0.040, 0.413],  $P=0.02$ ) (Table 2), and hsCRP was the primary contributing factor to plaque enhancement in participants <60 years (Figure 4).

## Discussion

To our knowledge, this is the first study to explore the age-varying associations between ICAS plaque enhancement and circulating inflammatory indicators. A positive correlation between hsCRP and ICAS plaque enhancement index on HR-MRI was observed, especially for participants <60 years. Moreover, the hsCRP level was the primary contributing factor to plaque enhancement in participants <60 years.

The ICAS plaque enhancement index is considered an important characteristic of vulnerable plaques, which may reflect active local inflammatory response and high risk of following stroke.<sup>11,22,23</sup> Histological studies have



**Figure 3** The scatter plot of circulating inflammatory indicators and ICAS plaque enhancement index for all participants, participants <60 years and participants  $\geq 60$  years. **Notes:** (A1-A3) Scatter plot of hsCRP and enhancement index for all participants, participants <60 years, and participants  $\geq 60$  years, respectively. (B1-B3) Scatter plot of LWR and enhancement index for all participants, participants <60 years, and participants  $\geq 60$  years, respectively. (C1-C3) Scatter plot of SII and enhancement index for all participants, participants <60 years, and participants  $\geq 60$  years, respectively. (D1-D3) Scatter plot of PLR and enhancement index for all participants, participants <60 years, and participants  $\geq 60$  years, respectively. (E1-E3) Scatter plot of LMR and enhancement index for all participants, participants <60 years, and participants  $\geq 60$  years, respectively. (F1-F3) Scatter plot of NLR and enhancement index for all participants, participants <60 years, and participants  $\geq 60$  years, respectively. Variables with skewed distributions were logarithmically converted for analysis.

**Abbreviations:** hsCRP, high-sensitivity C-reactive protein; LWR, lymphocyte-to-white blood cell ratio; SII, systemic immune inflammation index; PLR, platelet-to-lymphocyte ratio; LMR, lymphocyte-to-monocyte ratio; NLR, neutrophil-to-lymphocyte ratio.

**Table 2** Association Between Inflammatory Indicators and Plaque Enhancement Index

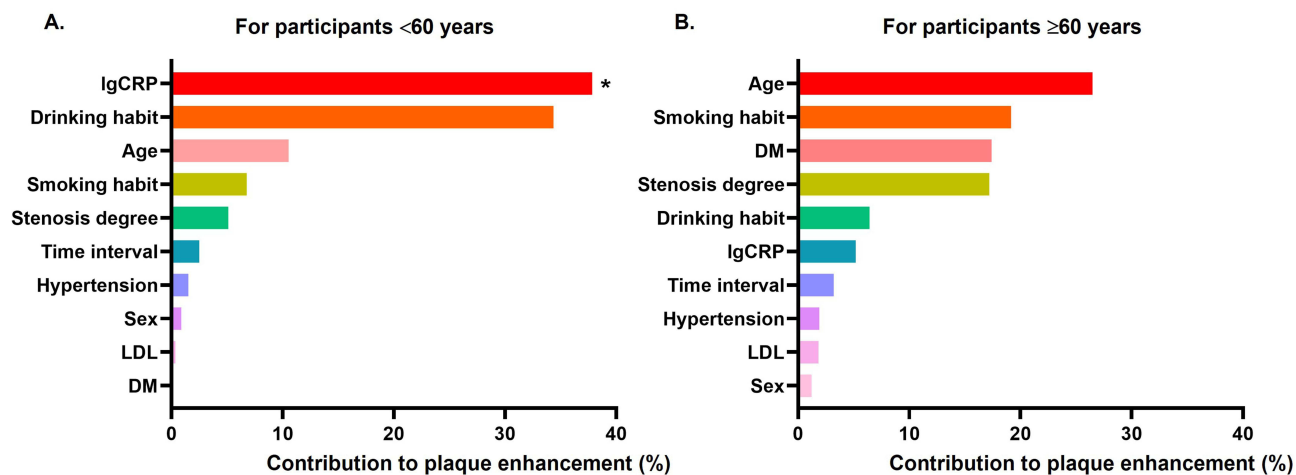
	Univariate regression		Multivariate regression					
			Model 1		Model 2		Model 3	
	$\beta$ (95% CI)	P	$\beta$ (95% CI)	P	$\beta$ (95% CI)	P	$\beta$ (95% CI)	P
For all participants								
hsCRP	0.219 (0.036, 0.349)	0.02	0.236 (0.043, 0.371)	0.01	0.222 (0.025, 0.364)	0.03	0.190 (-0.002, 0.337)	0.05
LWR	-0.018 (-0.594, 0.486)	0.85	-0.017 (-0.607, 0.507)	0.86	-0.028 (-0.656, 0.490)	0.78	0.001 (-0.564, 0.570)	0.99
SII	0.049 (-0.220, 0.382)	0.60	0.049 (-0.224, 0.385)	0.60	0.067 (-0.202, 0.425)	0.48	0.023 (-0.276, 0.352)	0.81
PLR	-0.002 (-0.508, 0.498)	0.99	-0.001 (-0.511, 0.506)	0.99	0.022 (-0.470, 0.591)	0.82	0.015 (-0.479, 0.562)	0.87
LMR	-0.051 (-0.569, 0.320)	0.58	-0.053 (-0.602, 0.340)	0.58	-0.051 (-0.616, 0.367)	0.62	-0.012 (-0.518, 0.458)	0.90
NLR	0.075 (-0.193, 0.466)	0.41	0.076 (-0.198, 0.472)	0.42	0.094 (-0.178, 0.518)	0.34	-0.046 (-0.267, 0.433)	0.64
For participants <60 years								
hsCRP	0.358 (0.055, 0.371)	0.01	0.340 (0.044, 0.361)	0.01	0.390 (0.057, 0.408)	0.01	0.381 (0.040, 0.413)	0.02
LWR	0.028 (-0.519, 0.632)	0.84	-0.013 (-0.600, 0.548)	0.93	-0.033 (-0.744, 0.611)	0.84	-0.033 (-0.744, 0.611)	0.84
SII	0.137 (-0.143, 0.413)	0.33	0.187 (-0.089, 0.459)	0.18	0.194 (-0.124, 0.507)	0.23	0.194 (-0.124, 0.507)	0.23
PLR	-0.123 (-0.727, 0.285)	0.38	-0.075 (-0.647, 0.378)	0.60	-0.059 (-0.700, 0.487)	0.72	-0.059 (-0.700, 0.487)	0.72
LMR	-0.021 (-0.514, 0.443)	0.88	-0.062 (-0.595, 0.387)	0.67	-0.063 (-0.670, 0.456)	0.70	-0.063 (-0.670, 0.456)	0.70
NLR	0.163 (-0.130, 0.491)	0.25	0.208 (-0.074, 0.536)	0.134	0.206 (-0.128, 0.583)	0.203	0.206 (-0.128, 0.593)	0.203
For participants $\geq 60$ years								
hsCRP	0.202 (-0.039, 0.447)	0.10	0.186 (-0.069, 0.445)	0.15	0.117 (-0.158, 0.395)	0.40	0.089 (-0.199, 0.378)	0.54
LWR	-0.047 (-0.985, 0.667)	0.70	-0.002 (-0.872, 0.861)	0.99	0.060 (-0.717, 1.118)	0.66	0.060 (-0.717, 1.118)	0.66
SII	0.028 (-0.453, 0.569)	0.82	-0.021 (-0.573, 0.485)	0.87	-0.073 (-0.696, 0.394)	0.58	-0.073 (-0.696, 0.394)	0.58
PLR	0.070 (-0.574, 1.029)	0.57	0.035 (-0.703, 0.932)	0.78	0.013 (-0.796, 0.883)	0.92	0.013 (-0.796, 0.883)	0.92
LMR	-0.084 (-0.913, 0.447)	0.50	-0.044 (-0.844, 0.597)	0.73	0.044 (-0.677, 0.920)	0.76	0.044 (-0.677, 0.920)	0.76
NLR	0.063 (-0.408, 0.690)	0.61	0.021 (-0.523, 0.618)	0.87	-0.036 (-0.690, 0.528)	0.79	-0.036 (-0.690, 0.528)	0.79

**Notes:** Model 1 was adjusted for age, sex. Model 2 was adjusted for age, sex, smoking habit, drinking habit, hypertension, DM, and LDL-c. Model 3 was adjusted for age, sex, smoking habit, drinking habit, hypertension, DM, LDL-c, stenosis degree, and time interval between HR-MRI and stroke.

**Abbreviations:** DM, diabetes mellitus; LDL-c, low-density lipoprotein cholesterol; hsCRP, high-sensitivity C-reactive protein; LWR, lymphocyte-to-white blood cell ratio; SII, systemic immune inflammation index; PLR, platelet-to-lymphocyte ratio; LMR, lymphocyte-to-monocyte ratio; NLR, neutrophil-to-lymphocyte ratio.

demonstrated that active inflammation is an important characteristic of vulnerable plaques.<sup>24</sup> During the progression of atherosclerosis, inflammation has been demonstrated to be involved in atherosclerotic thrombosis, playing a crucial role in plaque rupture, vascular embolization, and infarction.<sup>24,25</sup> Previous studies have shown that hsCRP is a sensitive indicator of circulating inflammatory response in atherosclerosis and plays a significant role in the pathogenesis of atherosclerosis.<sup>26,27</sup> High level of hsCRP was not only associated with ICAS but also with ICAS plaque enhancement.<sup>15,28</sup> Consistent with previous studies, we also found a positive correlation between hsCRP and ICAS plaque enhancement index. C-reactive protein can bind to lipoproteins, activate the complement system, produce a myriad of inflammatory mediators, release oxygen free radicals, leading to vascular intimal damage, vasospasm, and plaque rupture.<sup>29</sup> This association was believed to reflect the mutual influence between localized inflammation within the ICAS plaque and systemic inflammation. While no other circulating inflammation indicators, including LWR, SII, PLR, LMR, and NLR, were associated with ICAS plaque enhancement index. These novel circulating inflammation indicators are composite metrics derived from the proportions of various types of blood cells. Although these indicators can provide a more comprehensive reflection of the body's immune status and inflammatory response, they may lack specificity regarding inflammation associated with atherosclerosis, resulting in a failure to obtain correlations with ICAS plaque enhancement index in this study.

For the first time, we found that the association between hsCRP and ICAS plaque enhancement index was only significant in participants <60 years rather than participants  $\geq 60$  years. Meanwhile, hsCRP was the primary contributing factor to plaque enhancement in participants <60 years, whereas for participants  $\geq 60$  years, age had a more profound



**Figure 4** Contribution of hsCRP and other covariates to ICAS plaque enhancement in different age groups.

**Notes:** (A) For participants <60 years; (B) For participants ≥60 years. Age, sex, hypertension, DM, LDL-c, smoking habit, drinking habit, stenosis degree, and time interval between HRMRI and stroke were defined as covariates for analysis. \*Significant independent correlation in the regression model.

**Abbreviations:** hsCRP, high-sensitivity C-reactive protein; ICAS, intracranial atherosclerosis stenosis; DM, diabetes mellitus; LDL-c, Low-density lipoprotein cholesterol.

effect than hsCRP. Age is considered the strongest risk factor for atherosclerosis.<sup>30</sup> The pathological features of ICAS generally change from intimal thickening, fibrosis, and hyalinization to necrosis with aging.<sup>17</sup> ICAS generally progress slowly before the age of 60 years old and accelerates significantly after 60 years old. The aging process in humans is considered non-linear, with the age of 60 marking a significant turning point. Aging is associated with a decline in overall function, increasingly prevalent chronic diseases, persistent activation of the immune system, and high levels of systemic inflammation.<sup>31,32</sup> After reaching the age of 60 years old, the immune regulatory function declines more rapidly, oxidative stress responses become increasingly pronounced, and the incidence of vascular diseases rises notably.<sup>33</sup> This is particularly evident in the case of ICAS, where the progression significantly intensifies post 60 years old due to reduced activity of specific enzymes.<sup>17</sup> Before the age of 60, systemic inflammatory responses tend to remain at lower levels, making hsCRP, as a classic circulating inflammatory marker, more effective in explaining ICAS plaque enhancement. While after the age of 60 years old, ICAS plaque enhancement is largely attributed to the aging related systemic immune-inflammatory responses. Interestingly, hsCRP may not accurately capture the changes of systemic immune-inflammatory responses associated with aging. Besides, there were no significant differences between participants aged <60 years and ≥60 years in terms of stenosis degree, plaque burden, plaque enhancement index, and time interval between HR-MRI and stroke. Therefore, it was challenging to attribute the discrepancies in the association between hsCRP levels and plaque enhancement at different ages to plaque characteristics. According to previous studies, joint analysis of hsCRP and plaque enhancement index can better evaluate vulnerable plaques and the risk of future vascular events.<sup>15,16</sup> Through age stratification, we gained further insight into the relationship between ICAS plaque enhancement and systematic inflammation, which may help us better understand the significance of plaque enhancement in clinical practice which deserves further study.

This study should be interpreted with the following limitations. Firstly, the time interval between HR-MRI and stroke was an important factor influencing plaque enhancement.<sup>34,35</sup> Participants with different time intervals between HR-MRI and stroke were all included in this study and treated as a covariate for model adjustment. More stringent time limits are required for future studies to obtain a more precise understanding of the association between ICAS plaque enhancement and inflammation. Secondly, although we observed different associations between ICAS plaque enhancement and hsCRP across different age groups, no statistically significant interaction effect was observed between age and hsCRP. This may largely be limited by the sample size of the study, which is also confined to the Asian population. Further research is needed to solidify the conclusions and verify their applicability to other populations. Thirdly, other circulating inflammatory markers such as Interleukin-6 (IL-6) was not included in this study for analysis, which deserves further exploration.



## Conclusions

The ICAS plaque enhancement index was positively associated with hsCRP levels, particularly in participants aged <60 years. This may be helpful for understanding the significance of the enhancement index in clinical practice.

## Data Sharing Statement

The raw data supporting the conclusions of this study are made available by the corresponding author.

## Ethics Statement

This study was approved by the Ethical Standards Committee on Human Experimentation of Shandong Provincial Hospital, Shandong University, China. Written informed consent was obtained from all participants. This study was conducted in accordance with the ethical principles for medical research involving human subjects as expressed in the Declaration of Helsinki. Each participant signed an informed consent form after being informed of the purpose of the study.

## Acknowledgments

We are very grateful to all the participants in this study, the staff involved in the hospital, and the Steering Committee Members of this study.

## Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

## Funding

This study was supported by grants from the Department of Science and Technology of Shandong Province [ZR2020QH109 and ZR2022LSW010] and National Natural Science Foundation of China [82201477]. The funding agency had no role in the study design, data collection and analysis, writing of this article, or the decision to submit the manuscript for publication.

## Disclosure

The authors report no conflicts of interest in this work.

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## References

1. Feigin VL, Brainin M, Norrving B, et al. World Stroke Organization (WSO): global Stroke Fact Sheet 2022. *Int J Stroke*. 2022;17(1):18–29. doi:10.1177/17474930211065917
2. Paul S, Candelario-Jalil E. Emerging neuroprotective strategies for the treatment of ischemic stroke: an overview of clinical and preclinical studies. *Exp Neurol*. 2021;335:113518. doi:10.1016/j.expneurol.2020.113518
3. Qureshi AI, Caplan LR. Intracranial atherosclerosis. *Lancet*. 2014;383(9921):984–998. doi:10.1016/S0140-6736(13)61088-0
4. Ritz K, Denswil NP, Stam OC, van Lieshout JJ, Daemen MJ. Cause and mechanisms of intracranial atherosclerosis. *Circulation*. 2014;130(16):1407–1414. doi:10.1161/CIRCULATIONAHA.114.011147
5. Elhfnawy AM, Heuschmann PU, Pham M, Volkmann J, Fluri F. Stenosis length and degree interact with the risk of cerebrovascular events related to internal carotid artery stenosis. *Front Neurol*. 2019;10:317. doi:10.3389/fneur.2019.00317
6. Li GW, Zheng GY, Li JG, Sun XD. Relationship between carotid atherosclerosis and cerebral infarction. *Chin Med Sci J*. 2010;25(1):32–37. doi:10.1016/S1001-9294(10)60017-X
7. Gao T, Zhang Z, Yu W, Zhang Z, Wang Y. Atherosclerotic carotid vulnerable plaque and subsequent stroke: a high-resolution mri study. *Cerebrovasc Dis*. 2009;27(4):345–352. doi:10.1159/000202011
8. Millon A, Boussel L, Brevet M, et al. Clinical and histological significance of gadolinium enhancement in carotid atherosclerotic plaque. *Stroke*. 2012;43(11):3023–3028. doi:10.1161/STROKEAHA.112.662692
9. Lee HN, Ryu CW, Yun SJ. Vessel-wall magnetic resonance imaging of intracranial atherosclerotic plaque and ischemic stroke: a systematic review and meta-analysis. *Front Neurol*. 2018;9:1032. doi:10.3389/fneur.2018.01032

10. Kim JM, Jung KH, Sohn CH, et al. Intracranial plaque enhancement from high resolution vessel wall magnetic resonance imaging predicts stroke recurrence. *Int J Stroke*. 2016;11(2):171–179. doi:10.1177/1747493015609775
11. Lv YD, Ma XT, Zhao WH, et al. Association of plaque characteristics with long-term stroke recurrence in patients with intracranial atherosclerotic disease: a 3D high-resolution MRI-based cohort study. *Eur Radiol*. 2023;34(5):3022–3031. doi:10.1007/s00330-023-10278-y
12. Hur J, Park J, Kim YJ, et al. Use of contrast enhancement and high-resolution 3d black-blood mri to identify inflammation in atherosclerosis. *Cardiovasc Imaging*. 2010;3(11):1127–1135.
13. Wilson AM, Ryan MC, Boyle AJ. The novel role of c-reactive protein in cardiovascular disease: risk marker or pathogen. *Int J Cardiol*. 2006;106(3):291–297. doi:10.1016/j.ijcard.2005.01.068
14. Fani L, van Dam-Nolen DHK, Vernooij M, Kavousi M, van der Lugt A, Bos D. Circulatory markers of immunity and carotid atherosclerotic plaque. *Atherosclerosis*. 2021;325:69–74. doi:10.1016/j.atherosclerosis.2021.03.040
15. Li RY, Zhao DL, Yu JW, et al. Ju. Intracranial plaque characteristics on high-resolution MRI and high-sensitivity C-reactive protein levels: association and clinical relevance in acute cerebral infarction. *Clin Radiol*. 2023;78(5). doi:10.1016/j.crad.2023.01.004
16. Schwartz RS, Bayes-Genis A, Lesser JR, et al. Detecting vulnerable plaque using peripheral blood: inflammatory and cellular markers. *J Interv Cardiol*. 2003;16(3). doi:10.1034/j.1600-0854.2003.8025.x
17. Wang Y, Meng R, Liu G, et al. Intracranial atherosclerotic disease. *Neurobiol Dis*. 2019;124:118–132. doi:10.1016/j.nbd.2018.11.008
18. Qiao Y, Zeiler SR, Mirbagheri S, et al. Intracranial plaque enhancement in patients with cerebrovascular events on high-spatial-resolution MR images. *Radiology*. 2014;271(2):534–542. doi:10.1148/radiol.13122812
19. Lou X, Ma N, Ma L, Jiang WJ. Contrast-enhanced 3t high-resolution mr imaging in symptomatic atherosclerotic basilar artery stenosis. *AJNR Am J Neuroradiol*. 2013;34(3):513–517. doi:10.3174/ajnr.A3241
20. Wang M, Wu F, Yang Y, et al. Quantitative assessment of symptomatic intracranial atherosclerosis and lenticulostriate arteries in recent stroke patients using whole-brain high-resolution cardiovascular magnetic resonance imaging. *J Cardiovasc Magn Reson*. 2018;20(1):35. doi:10.1186/s12968-018-0465-8
21. Chung JW, Hwang J, Lee MJ, Cha J, Bang OY. Previous statin use and high resolution magnetic resonance imaging characteristics of intracranial atherosclerotic plaque: the intensive statin treatment in acute ischemic stroke patients with intracranial atherosclerosis study. *Stroke*. 2016;47(7):1789–1796. doi:10.1161/STROKEAHA.116.013495
22. Wang E, Shao S, Li S, et al. A high-resolution mri study of the relationship between plaque enhancement and ischemic stroke events in patients with intracranial atherosclerotic stenosis. *Front Neurol*. 2019;9:1154. doi:10.3389/fneur.2018.01154
23. Skarpathiotakis M, Mandell DM, Swartz RH, Tomlinson G, Mikulis DJ. Intracranial atherosclerotic plaque enhancement in patients with ischemic stroke. *AJNR Am J Neuroradiol*. 2013;34(2):299–304. doi:10.3174/ajnr.A3209
24. Pelisek J, Eckstein HH, Zerneck A. Pathophysiological mechanisms of carotid plaque vulnerability: impact on ischemic stroke. *Arch Immunol Ther Exp (Warsz)*. 2012;60(6):431–442. doi:10.1007/s00005-012-0192-z
25. Ross R. Atherosclerosis—an inflammatory disease. *N Engl J Med*. 1999;340(2):115–126. doi:10.1056/NEJM199901143400207
26. Badimon L, Peña E, Arderiu G, et al. C-reactive protein in atherothrombosis and angiogenesis. *Front Immunol*. 2018;9:430. doi:10.3389/fimmu.2018.00430
27. Denegri A, Boriani G. High sensitivity c-reactive protein (hsCRP) and its implications in cardiovascular outcomes. *Curr Pharm Des*. 2021;27(2):263–275. doi:10.2174/1381612826666200717090334
28. Su BJ, Dong Y, Tan CC, et al. Elevated hs-crp levels are associated with higher risk of intracranial arterial stenosis. *Neurotox Res*. 2020;37(2):425–432. doi:10.1007/s12640-019-00108-9
29. David S, Sridevi D, Mitra A, et al. Inflammation, atherosclerosis, and psoriasis. *Clin Rev Allergy Immunol*. 2013;44(2):194–204. doi:10.1007/s12016-012-8308-0
30. Peter L. The changing landscape of atherosclerosis. *Nature*. 2021;592:7855:524–533.
31. Culig L, Chu X, Bohr VA. Neurogenesis in aging and age-related neurodegenerative diseases. *Ageing Res Rev*. 2022;78:101636. doi:10.1016/j.arr.2022.101636
32. Amit S, Schurman Shepherd H, Arsun B, et al. Aging and Inflammation. *Cold Spring Harb Perspect Med*. 2024;14(6):a041197. doi:10.1101/cshperspect.a041197
33. Shen XT, Wang CC, Zhou X, et al. Nonlinear dynamics of multi-omics profiles during human aging. *Nat Aging*. 2024;4(1):1–6. doi:10.1038/s43587-023-00542-7
34. Zhang XF, Chen LG, Li S, et al. Enhancement characteristics of middle cerebral arterial atherosclerotic plaques over time and their correlation with stroke recurrence. *J Magn Reson Imaging*. 2021;53(3):953–962. doi:10.1002/jmri.27351
35. Yang WJ, Jill A, Yannie Oi-Yan S, et al. Regression of plaque enhancement within symptomatic middle cerebral artery atherosclerosis: a high-resolution MRI study. *Front Neurol*. 2020;11:755. doi:10.3389/fneur.2020.00755