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# Relationship of cerebral microbleeds to inflammatory marker levels

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## ABSTRACT

**Aim:** The purpose of this study is to investigate the incidence, distribution and risk factors of cerebral microbleeds (CMBs) and the relation between CMBs and inflammation in ischemic cerebrovascular disease. **Methods:** Two hundred and one patients without acute infarction or transient ischemic attack were enrolled. The presence and number of CMB were assessed on susceptibility-weighted imaging. The traditional risk factors of CMB were recorded. Levels of high-sensitivity C-reactive protein (hs-CRP), interleukin-6 (IL-6), and matrix metalloproteinase-9 (MMP-9) were tested. Logistic regression analyses were used for multiple-factor analysis of risk factors of CMB. **Results:** Of the 201 patients, 49 (24.38%) had CMB. Multivariate logistic regression analyses showed that the age, the prevalence of hypertension, silent lacunar infarction, white matter lesion, Montreal Cognitive Assessment Score, the using rate of antithrombotic drugs and levels of hs-CRP, IL-6, MMP-9 were the risk factors for CMB. After adjustments for traditional risk factors, inflammatory marker levels remained to be associated with CMBs. The adjusted odd ratios of hs-CRP, IL-6 and MMP-9 were 1.745 (1.342-2.270), 1.223 (1.018-1.533) and 1.284 (1.082-1.423), respectively. Furthermore, inflammatory marker levels were the risk factor for deep or infratentorial CMBs and lobar CMBs. **Conclusion:** The age, prevalence of hypertension, silent lacunar infarction, white matter lesion, MoCA Score, the using rate of antithrombotic drugs and serum hs-CRP, IL-6, and MMP-9 levels were the independent risk factors for CMBs.

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## INTRODUCTION

Cerebral microbleeds (CMBs) are designated as a microvascular disease which has no related symptoms and physical signs.[1] CMBs has a round or ovoid signal of hypointensity (d < 10 mm) in T2* Gradient-Recall Echo (GRE) and susceptibility-weighted imaging (SWI) sequences.[2] Previous studies suggested that damage of cerebral microvasculature resulted from hypertensive vasculopathy, cerebral amyloid angiopathy (CAA) and ischemic brain damage.[3] CMBs are associated with hypertensive vasculopathy tends to occur in basal ganglia, whereas advanced CAA is associated with a lobar distribution.[1] CMBs exists widely in patients of ischemic and hemorrhagic stroke, but also in normal elderly individuals.[1] Previous studies have shown that stroke patients combined with CMBs have a significantly higher risk of recurrence.[3] The presence of CMBs was also independently associated with cognitive functions.[4] Multiple CMBs always indicate serious microvascular lesion which will increase risk of rebleeding.[5] At the same time, patients with CMBs had a higher risk of spontaneous hemorrhage while receiving anticoagulation and thrombolytic therapy.[5] CMBs are related to antiplatelet-related intracerebral hemorrhage.[6] Recently, an individual participant meta-analysis showed that C-reactive protein concentration was associated with the risk of coronary heart disease and ischemic stroke.[7] Inflammatory marker interleukin-6 (IL-6) retained a significant association with coronary heart disease and ischemic stroke.[8] A review revealed that matrix metalloproteinase-9 (MMP-9) levels were significantly correlated with severity of stroke, larger infarct volume and worse functional outcome.[9] For the above reason, we sought to choose high-sensitivity C-reactive protein (hs-CRP), IL-6 and MMP-9 to investigate the relationship between inflammatory marker levels and CMBs so as to prevent CMBs from developing.

## METHODS

### Population

Between June 2013 and March 2015, we recruited 286 participants with a age was greater than 45 years of age from both outpatient and in-patient services of the Department of Neurology, Peking University Binhai Hospital. In this study, the majority of the patients were without acute or transient neurologic impairment. We excluded 18 patients with a history of intracerebral hemorrhage and 9 patients with a history of traumatic brain injury. In addition, infectious diseases (*n* = 12), collagen disease (*n* = 8), malignant disease (*n* = 4) and patients with severe co-morbid medical conditions (*n* = 10) and those using anti-inflammatory medications (*n* = 13) were also excluded. We further excluded patients whose hs-CRP levels were higher than 8 mg/L (*n* = 11). Finally, 201 patients were included in the analysis. We divided the patients into 2 groups based on SWI, CMBs group (*n* = 49) and no CMBs group (*n* = 152), CMBs group was further divided into deep or infratentorial CMBs group (*n* = 30) and lobar CMBs group (*n* = 19) based on CMBs location.

### Risk factors assessment

The data including age, gender, body weight, height and medical history of patients such as smoking, alcohol intake, heart disease，blood pressure, blood glucose, serum lipid and the use of antithrombotic drugs were recorded for analysis. Hypertension was defined as blood pressure ≥ 140/90 mmHg on measurements taken on at least two occasions, or patients with a history of hypertension and using antihypertensive drug. Hyperlipidemia was defined as triglyceride level ≥ 1.7 mmol/L; low-density lipoprotein cholesterol level ≥ 3.4 mmol/L; total cholesterol level ≥ 5.72 mmol/L, or use of cholesterol-lowering therapy. Diabetes was defined as fasting blood-glucose ≥ 7.0 mol/L and/or postprandial blood sugar ≥ 11.1 mol/L, or use of antidiabetic therapy. Body mass index was defined as weight [kg]/height [m]$^2$. Smoking was defined as ever smoking but giving up and current smoking. Habitual alcohol intake was defined as alcohol drinking more than 20 g/day antiplatelet drugs and anticoagulant drugs are regarded as antithrombotic drugs. Montreal Cognitive Assessment Scores were used to assess cognitive function.

### Magnetic resonance imaging parameters and diagnosis of CMB

Brain magnetic resonance imaging (MRI) was performed with a 3.0-tesla MR unit (Achieva; Philips). Susceptibility-weighted imaging (TR = 17 ms, TE = 24 ms, 10-mm-thick slices with 5-mm spacing) was included in the routine protocol. All the images were compared and analyzed on the workstation by trained and experienced radiologist and neurologist who were blinded to clinical information at the same time according to the Microbleed Anatomical Rating Scale MARS.[10] A single CMB was defined as a small, round, or ovoid hypointensity of < 10 mm in diameter, evident on T2* GRE or SWI MRI sequences.[2] Microbleed mimics (e.g. vessels, mineralization, air-bone interfaces, partial volume artifact at the edges of the cerebellum) were excluded.[10] CMBs were divided into lobar microbleeds (cortex, subcortex, and white matter) and deep or infratentorial

microbleeds (basal ganglia, thalamus, brainstem and cerebellum).[11] Further, we made categories for "strictly lobar microbleeds" (persons with ≥ 1 new microbleeds restricted to a lobar location) and "deep or infratentorial microbleeds" (persons with ≥ 1 new microbleeds in a deep or infratentorial location with or without lobar microbleeds).[12] Silent lacunar infarction was defined as an area of low signal intensity on T1-weighted images with corresponding high signal intensity on T2-weighted images, whose diameter was > 3 mm and < 15 mm (e.g. dilated perivascular space).[13] Diagnostic criteria of white matter lesion: periventricular white matter lesions (WMLs) were scored according to the following patterns: no lesions (0 points); pencil-like or cap-like thin lesions (1 point); smooth haloes at lesion site (2 points); and irregular periventricular high signals extending to deep WM (3 points). Deep WMLs were scored according to the following patterns: no lesions (0 points); punctate separate lesions (1 point); fused lesions (2 points); and large fused lesions (3 points).The total score was obtained by adding the periventricular and deep WMLs scales together.[14]

## Measurement of inflammatory markers

Blood was drawn with minimally traumatic venipuncture for measurement of serum inflammatory markers. Blood were centrifuged by 3,000 *g* for 15 min at 4 °C, and then aliquots were stored at -70 °C. Serum hs-CRP was measured by latex turbidimetric immunoassay with a sensitivity of 0.01 mg/L. Serum IL-6 and MMP-9 were measured by enzyme-linked immunosorbent assay (High Sensitivity Quantikine kit; R&D System) with a sensitivity of 0.01 pg/mL. Serum MMP-9 was also measured by enzyme-linked immunosorbent assay (High Sensitivity Quantikine

kit; R&D System). The detectable limit for Serum IL-6 and MMP-9 were 0.01 pg/mL and 0.01 ng/mL, respectively.

## Statistical analysis

Data was analyzed using SPSS 19.0. Measurement data was described as mean ± standard deviation (SD). Enumeration data was described as number (%). *T*-test and one-way analysis of variance was used for comparisons of continuous variables. We used $\chi^2$ test for enumeration data. Skewed distribution data was described as (Media and Q1-Q3). Kruskal-Wallis test followed by the Mann-Whitney *U* test were used for comparisons between groups. Multivariate logistic regression analyses were used for calculation of odds ratio, in which logarithmically transformed values of inflammatory markers were used. The results are shown as the odd ratios (OR) with 95% confidence interval (CI). Probability values were 2-tailed, and values of *P* < 0.05 were considered significant.

## RESULTS

### CMB distributional characteristics

Of the patients, 49 (24.38%) had CMBs. The spatial distribution of the CMB number and location was as follows: deep or infratentorial, 166 in 30 patients (61.22%); lobar, 88 in 19 patients (38.78%).

### Relations between CMBs and traditional risk factors

The baseline characteristics of the patients in this study are shown in Tables 1 and 2. There were significant differences in the traditional risk factors such as age, the prevalence of hypertension, coronary heart disease, silent lacunar infarction (SLI) and WMLs,

**Table 1: Comparison of baseline characteristics between CMB group and no CMB group**

| Characteristics | CMB group (*n* = 49) | No CMB group (*n* = 152) | *P* |
|---|---|---|---|
| Age (years) | 68.61 ± 7.76[†] | 61.76 ± 11.06 | 0.011 |
| Male (*n*, %) | 29 (59.18) | 79 (51.97) | 0.483 |
| BMI (kg/m$^2$) | 26.89 ± 2.96 | 25.63 ± 2.76 | 0.068 |
| Smoking (*n*, %) | 21 (42.85) | 37 (47.37) | 0.642 |
| Alcohol intake (*n*, %) | 26 (53.06) | 74 (48.68) | 0.494 |
| Hypertension (*n*, %) | 33 (67.34)[‡] | 61 (40.13) | 0.001 |
| DM (*n*, %) | 20 (40.81) | 60 (39.47) | 0.868 |
| HLP (*n*, %) | 21 (42.85) | 57 (37.50) | 0.506 |
| CHD (*n*, %) | 29 (59.18)[†] | 64 (42.10) | 0.048 |
| SLI (*n*, %) | 35 (71.42)[‡] | 47 (30.92) | 0.001 |
| WML (M, Q1-Q3) | 5 (4-6)[‡] | 2 (1-4) | 0.000 |
| Antithrombotic drugs (*n*, %) | 27 (55.10)[†] | 54 (35.53) | 0.019 |
| MoCA Score | 24.45 ± 0.29[‡] | 26.62 ± 0.21 | 0.006 |
| hs-CRP [mg/L (M, Q1-Q3)] | 6.83 (5.91-9.73)[‡] | 3.20 (2.10-5.34) | 0.000 |
| IL-6 [pg/mL (M, Q1-Q3)] | 8.23 (6.68-12.20)[‡] | 5.59 (3.72-7.79) | 0.000 |
| MMP-9 [ng/mL (M, Q1-Q3)] | 15.98 (13.65-18.46)[‡] | 11.66 (7.78-15.77) | 0.001 |

[†]*P* < 0.05, [‡]*P* < 0.01, compared with the no CMB group. CMB: cerebral microbleeds; BMI: body mass index; DM: diabetes mellitus; HLP: hyperlipemia; CHD: coronary heart disease; SLI: silent lacunar infarction; hs-CRP: high-sensitivity C-reactive protein; IL: interleukin; MMP: matrix metalloproteinase

MoCA Scores or the using rate of antithrombotic drugs between the CMB group and no CMB group (*P* < 0.05, Table 1). Furthermore, we also observed similar associations between traditional risk factors and deep or infratentorial CMBs [Table 2]. Similarly, there were significant differences such as age, or the prevalence of alcohol intake, SLI and WML between the lobar CMB and no CMB group (*P* < 0.05, Table 2). Compared with the lobar CMB group, there was higher prevalence of hypertension and lower alcohol intake in deep or infratentorial CMB group [Table 2]. On logistic regression analysis, the association between traditional risk factors and CMBs are summarized in Table 3. The ORs (per 1SD increase; 95% CI) of age, the prevalence of hypertension, SLI and WML, MoCA Score and the using rate of antithrombotic drugs were 1.666 (1.062-2.632), 2.634 (1.067-4.767), 2.345 (1.056-5.226), 1.717 (1.132-2.603), 1.198 (1.023-2.268) and 1.234 (1.116-2.584), respectively. In addition, the association between traditional risk factors and deep or infratentorial CMBs were also stronger [Table 3]. However, for strictly lobar CMB, the only independent risk factor was age and the prevalence of alcohol intake [Table 3].

## Relations between CMBs and inflammatory marker levels

We observed that serum hs-CRP, IL-6 and MMP-9 levels (median: 6.83, 8.23, 15.98, respectively) in the CMB group were higher than in those (median: 3.20, 5.59, 11.66, respectively) without CMB group [Table 1]. Similarly, we observed that hs-CRP, IL-6 and MMP-9 levels were higher in the deep or infratentorial CMB group (median: 7.01, 9.31, 17.63, respectively) or lobar CMB group (median: 6.82, 7.98, 15.84, respectively) than in those without [Table 2]. There was no significant difference between the deep or infratentorial CMB group and lobar CMB group. Connections between CMB and inflammatory markers are summarized in Table 4. Logistic regression analysis showed that each 1SD-increase in each inflammatory marker level was significantly associated with the presence of CMB after adjustment for age and sex (Model 1). After adjustment for age, sex, BMI, smoking, alcohol intake, hypertension, diabetes, hyperlipidemia, coronary heart disease, the useful of antithrombotic drugs and the presence of SLI and WML, inflammatory marker level remained to be associated with CMB (Model 2).

The adjusted ORs of hs-CRP, IL-6 and MMP-9

### Table 2: Comparison of baseline characteristics between deep or infratentorial CMB group and lobar CMB group

| Characteristics | Deep or infratentorial CMB group (*n* = 30) | Lobar CMB group (*n* = 19) | No CMB group (*n* = 152) |
|---|---|---|---|
| Age (years) | 69.77 ± 7.76‡ | 67.54 ± 7.88‡ | 61.76 ± 11.06 |
| Male (*n*, %) | 19 (63.33) | 10 (52.63) | 79 (51.97) |
| BMI (kg/m$^2$) | 26.65 ± 3.35 | 25.39 ± 2.89 | 25.63 ± 2.76 |
| Smoking (*n*, %) | 11 (36.67) | 9 (47.37) | 37 (47.37) |
| Alcohol intake (*n*, %) | 12 (40.00)* | 14 (73.68)† | 74 (48.68) |
| Hypertension (*n*, %) | 24 (80.00)‡* | 9 (47.37) | 61 (40.13) |
| DM (*n*, %) | 13 (43.33) | 7 (36.84) | 60 (39.47) |
| HLP (*n*, %) | 14 (46.67) | 7 (36.84) | 57 (37.50) |
| CHD (*n*, %) | 20 (66.67)† | 10 (52.63) | 64 (42.10) |
| SLI (*n*, %) | 24 (80.00)‡ | 11 (57.89)† | 47 (30.92) |
| WML (M, Q1-Q3) | 5.5 (4-6)‡ | 5 (4-6)† | 2 (1-4) |
| Antithrombotic drugs (*n*, %) | 18 (60.00) † | 9 (47.37) | 54 (35.53) |
| MoCA Score | 24.20 ± 1.56‡ | 24.58 ± 1.07† | 26.62 ± 0.21 |
| hs-CRP [mg/L (M, Q1-Q3)] | 7.01 (5.23-9.93)‡ | 6.82 (5.67-9.80)‡ | 3.20 (2.10-5.34) |
| IL-6 [pg/mL (M, Q1-Q3)] | 9.31 (8.15-13.82)‡ | 7.98 (5.80-10.81)‡ | 5.59 (3.72-7.79) |
| MMP-9 [ng/mL (M, Q1-Q3)] | 17.63 (14.02-18.91)‡ | 15.84 (13.02-16.74)‡ | 11.66 (7.78-15.77) |

†*P* < 0.05, ‡*P* < 0.01, compared with the no CMB group; **P* < 0.05, ***P* < 0.01, compared with the lobar CMB group. CMB: cerebral microbleeds; BMI: body mass index; DM: diabetes mellitus; HLP: hyperlipemia; CHD: coronary heart disease; SLI: silent lacunar infarction; hs-CRP: high-sensitivity C-reactive protein; IL: interleukin; MMP: matrix metalloproteinase

### Table 3: OR (95% CI) for CMB status per 1SD increase in traditional risk factors

| Variable | CMB OR (95% CI) | Deep or infratentorial CMB OR (95% CI) | Lobar CMB OR (95% CI) |
|---|---|---|---|
| Age | 1.666 (1.062-2.632)† | 1.773 (1.172-2.641)† | 1.436 (1.134-2.442)† |
| Hypertension | 2.634 (1.067-4.767)† | 3.875 (1.795-5.955)‡ | |
| SLI | 2.345 (1.056-5.226)† | 2.438 (1.226-4.750)‡ | |
| WML | 1.717 (1.132-2.603)† | 1.850 (1.291-2.643)† | |
| MoCA Score | 1.198 (1.023-2.268)† | 1.230 (1.100-2.448)† | |
| Antithrombotic drugs | 1.234 (1.116-2.584)† | 1.290 (1.001-2.229)‡ | |
| Alcohol intake | | | 1.023 (1.008-1.058)† |

†*P* < 0.05, ‡*P* < 0.01, compared with the no CMB group. SD: standard deviation; CI: confidence interval; OR: odd ratios; CMB: cerebral microbleeds; SLI: silent lacunar infarction; WML: white matter lesion

**Table 4: OR (95% CI) for CMB status per 1SD increase in inflammation factors (values were log transformed for analysis)**

| Variable | CMB | | Deep or infratentorial CMB | | Lobar CMB | |
|---|---|---|---|---|---|---|
| | Model 1 OR (95% CI) | Model 2 OR (95% CI) | Model 1 OR (95% CI) | Model 2 OR (95% CI) | Model 1 OR (95% CI) | Model 2 OR (95% CI) |
| hs-CRP | 1.852 (1.435-2.391)‡ | 1.745 (1.342-2.270)‡ | 2.372 (1.680-3.262)‡ | 2.302 (1.520-3.482)‡ | 1.578 (1.043-2.337)‡ | 1.534 (1.009-2.259)‡ |
| IL-6 | 1.469 (1.204-1.792)† | 1.223 (1.018-1.533)‡ | 1.480 (1.102-2.492)‡ | 1.334 (1.008-3.660)† | 1.422 (1.001-2.678)‡ | 1.508 (1.097-3.428)† |
| MMP-9 | 1.397 (1.196-1.632)‡ | 1.284 (1.082-1.423)‡ | 1.420 (1.241-1.799)‡ | 1.287 (1.145-1.599)‡ | 1.293 (1.142-2.423)‡ | 1.242 (1.178-1.409)‡ |

†$P < 0.05$, ‡$P < 0.01$, compared with the no CMB group. Model 1: adjusted for age and sex; Model 2: adjusted for age, sex, body mass index, smoking, alcohol intake, hypertension, diabetes, hyperlipidemia, coronary heart disease, the presence of silent lacunar infarction and white matter lesion and antithrombotic drugs. CMB: cerebral microbleeds; SD: standard deviation; CI: confidence interval; OR: odd ratios; hs-CRP: high-sensitivity C-reactive protein; IL: interleukin; MMP: matrix metalloproteinase

were 1.745 (1.342-2.270), 1.223 (1.018-1.533) and 1.284 (1.082-1.423), respectively. Furthermore, the associations between inflammatory marker level and deep or infratentorial CMB or lobar CMB remained significant after adjustment for age and sex (Model 1) and after additional adjustment for the traditional risk factors (Model 2). The adjusted ORs of hs-CRP, IL-6 and MMP-9 for the presence of deep or infratentorial CMB were 2.302 (1.520-3.482), 1.334 (1.008-3.660) and 1.287 (1.145-1.599), respectively. For the presence of lobar CMB, the adjusted ORs of hs-CRP, IL-6 and MMP-9 were 1.534 (1.009-2.259), 1.508 (1.097-3.428) and 1.242 (1.178-1.409), respectively.

## DISCUSSION

The prevalence of CMBs varies from different population. A meta-analyze found that incidence of CMBs was 44% in ischemic stroke, 83% in intracerebral hemorrhage, and 5-6% in healthy adults.[15] In this study, the incidence of CMBs was 24.38%, 30 patients (61.22%) had deep or infratentorial CMB, 19 patients (38.78%) had lobar CMB. The prevalence of CMB in our study was higher than that in healthy adults, but, lower than that in ischemic stroke. The first possible reason was the patients who participated without acute infarction or transient ischemic attack. The second possible reason was the use of SWI, it may increase the prevalence and number of CMB detected, compared with T2 GRE.

On assessment, CMB are often categorized according to location, separating strictly lobar CMB from deep or infratentorial CMB. Lobar CMB represents cerebral amyloid angiopathy, but, deep and infratentorial CMB is attributed to hypertensive arteriopathy.[16] Poels *et al.*[17] found an association between atherosclerosis and deep or infratentorial CMB in individuals with uncontrolled hypertension. This may explain the theoretical assumption that deep or infratentorial CMB are attributed to hypertensive vasculopathy, whereas lobar CMB are a

result of amyloid angiopathy.[18] Several studies have demonstrated strictly that lobar CMB was associated with the apolipoprotein E4 allele and diastolic blood pressure. On the other hand, systolic blood pressure, pulse pressure, silent lacunar infarction, white matter hyperintensities and smoking were associated with CMB in the deep or infratentorial regions.[19] In our study, hypertension, SLI, WML were associated with higher incidence of CMB, and thus were strong risk factors of CMB. The associations between traditional risk factors and deep or infratentorial CMB remained significant. Our results were consistent with previous studies. Shams *et al.*[16] observed that patients with CMB were significantly aged, had hypertension, and had lower cognitive function. A number of studies have shown that age is an independent risk factor of CMB. Vernooij *et al.*[11] found that the incidence of CMB increased with age from 17.8% in persons aged 60-69 years to 38.3% in those over 80 years. There were significant differences in MoCA Scores and using antithrombotic drugs between the CMB group and no CMB group. Several studies reported an association between the presence of CMB and impaired cognitive function.[20,21] Small vessel disease can be visualized as white matter hyperintensities and SLI but also as CMB on brain MRI.[21] A number of studies found that almost all the CMB patients had various degrees of WML and different numbers of SLI. Gregoire *et al.*[6] found that CMB were more numerous and prevalent in antiplatelet users who developed symptomatic ICH compared with matched antiplatelet-users who did not develop ICH. This data suggested a potential role for CMB as a risk factor for antiplatelet-associated ICH. The relationship between alcohol intake and CMB needed to be further studied.

Our study showed that the levels of hs-CRP, IL-6, MMP-9 in CMB group were significantly higher than those in no CMB group. The regression analysis showed that inflammatory factors such as hs-CRP, IL-6 and MMP-9 were the independent risk factors of CMB. Also, they were also the independent risk factors of deep or infratentorial CMB and lobar CMB. This suggested that

CMB can be seen as the common downstream product of hypertensive vasculopathy and cerebral amyloid angiopathy pathways.[22] In old patients, hemosiderin deposited around the capillaries, the hemosiderin leaking through expansile vascular clearance would accelerate the inflammation and result in small vessel disease and CMB.[23] Previous studies found that levels of hs-CRP, IL-6, and IL-18 are related to both deep and lobar CMB.[22] Hoshi *et al.*[13] found that IL-6 could result in increase of hs-CRP as an important cytokine in inflammation, it can be a predictor of SLI. Koh *et al.*[24] found that the levels of MMP-9 and hs-CRP were significantly higher in patients with CMB than in those without. Hemosiderin of brain microbleed is well known to activate MMP-9, increased MMP-9 activity enhances inflammatory markers related to deterioration of neurological function in ischemic stroke.[24] Pantoni *et al.*[25] found that destruction of Blood Brain Barrie played an important role during the process of CMB. MMP-9 would destroy the blood-brain barrier (BBB) by degrading the extracellular matrix, leading to the increased permeability of BBB.[26] The animal experiment also proved a close relationship between increased MMP-9 and chronic destruction of BBB.[27] CMB resulting from cerebral amyloid angiopathy could be confirmed as the presence of β-amyloid in the vessel wall, which activated microglia and T lymphocytes expressing heme oxygenase-1 activity and complement activation were prominent. The above studies supported that inflammatory reaction participate the dysfunction of vascular endothelium and destruction of BBB, finally leading to the occurrence of CMB.[28]

This study has several limitations. Firstly, the results of this research have limited effectiveness because of the relatively small number of cases, especially deep or infratentorial and lobar CMB group. More patients must be enrolled in a large clinical trial. Secondly, in the cross-sectional study, we did not recruit the general population as a control group. The relationships of CMB and inflammation in the general population remain unclear. Thirdly, we choose hs-CRP, IL-6 and MMP-9 to study the relationship between inflammatory marker and CMB. Other inflammatory factors need to be studied further to confirm the relationship between inflammatory marker and CMB.

In conclusion, our results demonstrated that CMB were closely related with the age, prevalence of hypertension, SLI, WML, MoCA Score, the using rate of antithrombotic drugs, levels of hs-CRP, IL-6, and MMP-9, suggesting a role of inflammatory processes in CMB. Patients with CMB have been consistently found to have an elevated risk of hemorrhagic stroke or an antithrombotic-related hemorrhagic complication.[29] In the near future, we may reduce levels of circulating inflammatory markers to prevent CMB from happening.

## DECLARATIONS

### Authors' contributions
Substantially contributed to the general idea and design of the study: C. Li, Z.R. Jia
Took part in designing the protocol: Y. Song
Contributed to data collection and drafted the manuscript: Q.L. Lu
Helped with the data analysis: L. Wang
Read and approved the final manuscript: Q.L. Lu, C. Li, Y. Song, L. Wang, Z.R. Jia

### Conflicts of interest
The authors declare that they have no conflict of interest.

### Patient consent
Informed consent was obtained from all individual participants included in the study.

### Ethics approval
All procedures performed in the studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

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