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Hemoglobin level and three-month clinical outcomes among ischemic stroke patients with elevated systolic blood pressure

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ABSTRACT

Background: Previous studies have reported that extreme low and high hemoglobin levels are positively associated with the risk of ischemic stroke. However, there are few reports on the relationship between hemoglobin at acute phase and clinical outcomes after ischemic stroke and the results of their association to date are inconsistent. We aimed to investigate the association between them in a large prospective cohort of ischemic stroke patients.

Methods: Baseline hemoglobin levels were measured in 3881 patients with acute ischemic stroke. The primary outcome was defined as composite outcome of major disability and death (modified Rankin Scale score ≥ 3) at 3 months after stroke onset. Secondary outcomes were separately those of major disability and death.

Results: Compared with the lowest quartile of hemoglobin, the multivariate adjusted odds ratios (95% confidence intervals) associated with the highest quartile were 1.38 (1.03–1.86), 1.49 (1.11–1.99), 0.79 (0.41–1.52) for primary outcome, major disability and death, respectively. Multiple-adjusted spline regression model showed linear associations of hemoglobin levels with primary outcome (P for linearity = 0.037) and major disability (P for linearity = 0.004). Subgroup analyses further confirmed the positive association between high hemoglobin and poor prognosis of ischemic stroke.

Conclusions: Elevated hemoglobin levels in the acute phase were associated with poor prognosis at 3 months after ischemic stroke. Further prospective studies from other samples of ischemic stroke patients are needed to validate our findings.

1. Introduction

Stroke is the second leading cause of death and the most common cause of long-term disability in adults [1]. Hypoxia often happens during the first days after acute ischemic stroke onset and it has been reported to be able to result in secondary brain damage [2,3]. In order to increase the blood oxygen, supplemental oxygen is often used to improve prognosis in clinical treatment, but sometimes it may be harmful to patients [3]. Hemoglobin, a well-known iron-containing oxygen-transport protein [4], is commonly used in clinical practice as a marker of anemia (low hemoglobin) and vascular blood clotting (high hemoglobin) [5]. Previous studies demonstrated that hemoglobin concentrations could affect cardiovascular system through oxygen supply, blood viscosity and vasoconstriction [6,7]. Low hemoglobin levels have been shown to be able to increase the risk of cardiovascular death [8], and elevated hemoglobin levels are also associated with the risk of hypertension and carotid atherosclerosis [9–11]. In term of stroke, both lower and higher extremes of hemoglobin concentration were found to be positively associated with the incidence of ischemic stroke [12]. However, few studies investigating the association between hemoglobin

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and prognosis of ischemic stroke have yielded inconsistent results [13,14], which may be due to small size of sample, incomplete data or heterogeneous subjects simultaneously including ischemic stroke patients with thrombolytic therapy and those without thrombolytic therapy. Therefore, large-sample and well-designed prospective studies on the relationship between hemoglobin levels and clinical outcomes after ischemic stroke are needed. Given that high levels of hemoglobin can increase blood viscosity which may result in worse cardiovascular function, we hypothesize that elevated hemoglobin levels in the acute phase are associated with poor prognosis at 3 months after ischemic stroke. This study prospectively examined whether the hemoglobin level was independently associated with prognosis of ischemic stroke based on China Antihypertensive Trial in Acute Ischemic Stroke (CATIS).

2. Methods

2.1. Study patients

The present study was conducted among ischemic stroke patients from the CATIS study, a multicenter, single-blind, blinded end-points randomized clinical trial across China. Details of trial design, methods and major findings of the study have been described previously [15]. Briefly, 4071 ischemic stroke patients from 26 hospitals were recruited between August 2009 and May 2013 to examine whether immediate blood pressure (BP) reduction in patients with acute ischemic stroke would reduce death and major disability at 14 days or hospital discharge. The inclusion criteria of CATIS were to meet all of the followings: age of 22 years or older, having ischemic stroke confirmed by computed tomography or magnetic resonance imaging of the brain within 48 h of symptom onset, and having an elevated systolic BP between 140 mmHg and < 220 mmHg. Patients with a BP ≥ 220/120 mmHg, severe heart failure, acute myocardial infarction, unstable angina, atrial fibrillation, aortic dissection, cerebrovascular stenosis (≥70%), resistant hypertension, deep coma or treatment of intravenous thrombolytic therapy were excluded from the CATIS. In this study, 190 patients were further excluded because we lacked their baseline records of hemoglobin levels ($n = 94$) or they were lost to follow-up at 3 months ($n = 96$). Finally, a total of 3881 acute ischemic stroke patients were included in present analysis (Fig. 1).

This study was approved by the institutional review boards at Soochow University in China and Tulane University in the United States, as well as ethical committees at the participating hospitals. All study participants or their immediate family members agreed to take part in the study and provided written informed consents.

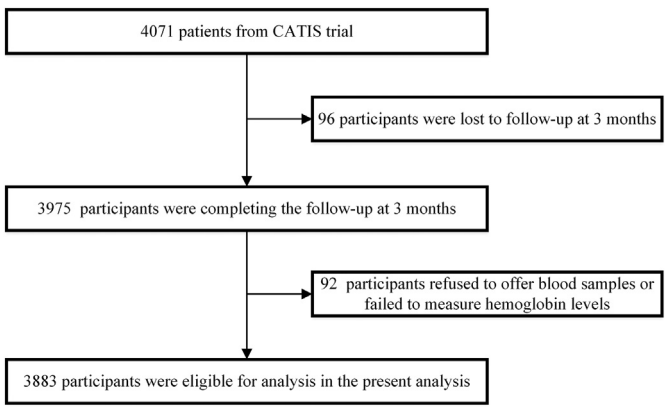

Fig. 1. Study participants flow chart.

2.2. Hemoglobin measurement

Blood samples were collected after at least 8 h of fasting within 24 h of hospital admission. Hemoglobin concentrations were measured for all enrolled patients in each participating hospital at admission.

2.3. Outcome assessment

The primary outcome of this study was a combination of death and major disability (modified Rankin Scale [mRS] score, 3–6) at 3 months after stroke onset [15–17]. The secondary outcomes separately those of major disability (mRS, 3–5) and death (mRS, 6). Death certificates were obtained for deceased patients. A trial-wide outcomes assessment committee, blinded to treatment assignment, reviewed and adjudicated subsequent outcomes based on the criteria established in the Antihypertensive and Lipid-Lowering Treatment to Prevent Heart Attack Trial.

2.4. Assessment of potential covariates

Demographic characteristics (age, sex, current cigarette smoking, current alcohol drinking), medical history, the time from onset to randomization and anthropometric indicators were collected at the time of enrollment [15]. According to the symptoms and imaging data of the patients, ischemic stroke were classified as large artery atherosclerosis (thrombotic), cardiac embolism (embolic) and small artery occlusion lacunae (lacunar) [18]. Stroke severity was estimated by trained neurologists at baseline using the National Institutes of Health Stroke Scale (NIHSS) [19]. Systolic and diastolic BP was defined as the average of three measurements when the patient was in the supine position using a standard mercury sphygmomanometer. Other clinical features including routine laboratory analyses (blood lipids, fasting plasma glucose, etc.) were performed at admission for all enrolled patients in each participating hospital.

2.5. Statistical analysis

Hemoglobin at baseline was categorized into four groups: Q1 (< 131 g/L), Q2 (131–142 g/L), Q3 (142–152 g/L) and Q4 (≥152 g/L) according to hemoglobin quartiles. Tests for linear trend of baseline characteristics across hemoglobin quartiles were performed using covariance analysis for continuous variables and chi-square trend analysis for categorical variables. Logistic regression analysis was used to estimate the association between hemoglobin level and poor clinical outcomes by calculating odds ratio (OR) and 95% confidence interval (CI) for higher quartiles compared to the lowest quartile. Trends for the ORs of ischemic stroke prognosis across increasing hemoglobin categories were determined, having hemoglobin category as an ordinal variable. We performed two multiple-adjusted logistic regression models. In the Model 1, the covariates included age, sex, time from onset to hospitalization, antihypertensive treatment, current smoking, alcohol consumption, body mass index, dyslipidemia, fasting plasma glucose and SBP at baseline, ischemic stroke subtypes, history of hypertension, diabetes and coronary heart disease, and family history of stroke. In the Model 2, we adjusted for the factors in Model 1 and further adjusted for baseline NIHSS score. In addition, spline regression models were used to test linearity assumption of association between hemoglobin and clinical outcomes, fitting a restricted cubic spline function with four knots (at the 5th, 35th, 65th, and 95th percentiles) [20]. We further assessed the potential effect modification by some stroke-related risk factors (i.e., age, sex, education, BMI, admission NIHSS score, current cigarette smoking, current alcohol drinking, history of hypertension, receiving immediate BP reduction). Interactions between hemoglobin and subgroup variables on the primary outcome were tested in the models with interaction terms by the likelihood ratio test, adjusting for the aforementioned covariates unless the variable was used as a

Table 1
Characteristics of participants according to hemoglobin quartile.

Characteristics	Hemoglobin, g/L					P_{trend}
	Total	Q1 (<131)	Q2 (131–142)	Q3 (142–152)	Q4 (≥152)	
Number of subjects	3883	910	990	986	997	
Demographic						
Age, years	62.5 ± 10.9	66.6 ± 11.2	64.4 ± 9.9	61.5 ± 10.2	57.8 ± 10.5	< 0.001
Male sex	2488 (64.1)	288 (31.7)	548 (55.4)	739 (75.0)	913 (91.6)	< 0.001
Current cigarette smoking	1440 (37.1)	171 (18.8)	334 (33.7)	413 (41.9)	522 (52.4)	< 0.001
Current alcohol drinking	1212 (31.2)	110 (12.1)	253 (25.6)	362 (36.7)	487 (48.9)	< 0.001
Clinical features						
Time from onset to randomization, h*	10.0 (4.5, 24.0)	12.0 (5.0, 24.0)	9.8 (4.0, 24.0)	10.0 (4.7, 24.0)	10.3 (5.0, 24.0)	0.689
Baseline systolic BP, mm Hg	166.1 ± 16.9	166.2 ± 16.7	165.6 ± 16.5	166.2 ± 17.2	166.5 ± 17.2	0.534
Baseline diastolic BP, mm Hg	96.7 ± 11.1	93.9 ± 10.9	95.7 ± 10.3	97.3 ± 11.1	99.5 ± 11.4	< 0.001
Body mass index, kg/m^2	25 ± 3.1	24.7 ± 3.4	24.9 ± 3.3	25.1 ± 2.9	25.3 ± 2.9	< 0.001
Dyslipidemia	2119 (54.6)	476 (52.3)	512 (51.7)	548 (55.6)	583 (58.5)	0.002
Baseline NIHSS score*	4.0 (2.0, 7.0)	5.0 (3.0, 8.0)	4.0 (2.0, 7.0)	4.0 (2.0, 7.0)	4.0 (2.0, 7.0)	0.017
Baseline fasting plasma glucose, mmol/L	6.7 ± 2.8	6.5 ± 2.6	6.6 ± 2.6	6.8 ± 2.9	6.9 ± 2.9	< 0.001
Medical history						
History of hypertension	3058 (78.8)	708 (77.8)	781 (78.9)	783 (79.4)	786 (78.8)	0.553
History of hyperlipidemia	268 (6.9)	49 (5.4)	70 (7.1)	66 (6.7)	83 (8.3)	0.022
History of diabetes mellitus	686 (17.7)	173 (19)	166 (16.8)	173 (17.6)	174 (17.5)	0.502
History of coronary heart disease	427 (11)	127 (14)	118 (11.9)	86 (8.7)	96 (9.6)	< 0.001
Family history of stroke	729 (18.8)	157 (17.3)	176 (17.8)	184 (18.7)	212 (21.3)	0.021
Ischemic stroke subtype						
Thrombotic	3028 (78)	701 (77)	776 (78.4)	774 (78.5)	777 (77.9)	0.655
Embolic	189 (4.9)	48 (5.3)	48 (4.9)	49 (5)	44 (4.4)	0.430
Lacunar	765 (19.7)	187 (20.6)	192 (19.4)	190 (19.3)	196 (19.7)	0.641
Receiving immediate BP reduction	1937 (49.9)	462 (50.8)	499 (50.4)	469 (47.6)	507 (50.9)	0.737

Abbreviations: BP = blood pressure; NIHSS = National Institute of Health Stroke Scale;
Data were presented as mean ± SD or n (%) unless otherwise noted;
* Data were presented as median (interquartile range).

subgroup variable. All *P* values were 2-tailed, and a significance level of 0.05 was used. Statistical analysis was conducted using SAS statistical software (version 9.4, Cary, North Carolina, USA).

3. Result

3.1. Baseline characteristics

A total of 3883 patients (2488 men and 1395 women; mean age, 62.5 ± 10.9 years) were included in the present study. The median hemoglobin concentration was 142.0 g/L (interquartile range, 131.0–152.0 g/L). The patients with higher hemoglobin were more likely to be younger, male, cigarette smokers, alcohol drinkers; to have higher admission diastolic BP, BMI, and fasting plasma glucose; to have a higher prevalence of dyslipidemia, history of hyperlipidemia and family history of stroke; to have lower NIHSS score; and to have lower prevalence of coronary heart disease history, compared with those with lower hemoglobin (Table 1).

3.2. Hemoglobin level and clinical outcomes

At 3-month follow-up visit, 979 (25.2%) patients experienced the primary outcome (860 major disabilities and 119 deaths). After adjustment for age, sex and other potential confounders in model 1, the adjusted OR of primary outcome associated with the highest quartile of hemoglobin was 1.34 (95% CI, 1.04–1.74; P_{trend} = 0.021). We further adjusted for baseline NIHSS score in model 2 and the adjusted OR of primary outcome for highest vs. lowest quartile of hemoglobin was 1.38 (95%CI, 1.03–1.86; P_{trend} = 0.043). Similar significant findings were observed for major disability in model 1 (OR: 1.44; 95%CI, 1.11–1.89; P_{trend} = 0.004) and model 2 (OR: 1.49; 95%CI, 1.11–1.99; P_{trend} = 0.006), respectively. However, there was no significant association of baseline hemoglobin with death within 3 months after stroke (Table 2).

We further used multivariable spline regression models to test linearity assumption of association between hemoglobin levels and 3-month clinical outcomes after stroke onset. As shown in Fig. 2A, there was a linear relationship between hemoglobin levels and 3-month primary outcome (*P* for linearity = 0.037). In addition, similar linear association of hemoglobin levels with major disability was observed among ischemic stroke patients (*P* for linearity = 0.004, Fig. 2B).

In subgroup analyses stratified by age, sex, education, BMI, admission NIHSS score, current cigarette smoking, current alcohol drinking, history of hypertension, receiving immediate BP reduction, the modest positive associations of hemoglobin with primary outcome and major disability were observed in almost all subgroups, and reached statistical significance in several subgroups. Moreover, no significant interaction was observed between hemoglobin levels and these interested factors in relation to primary outcome or major disability among ischemic stroke patients ($P_{interaction}$ > 0.05 for all, Fig. 3).

4. Discussion

In this prospective study based on CATIS, elevated hemoglobin levels at baseline were associated with 3-month poor prognosis after adjustment for conventional prognostic factors among 3883 ischemic stroke patients. The associations between them remained significant after further adjustment for NIHSS. Moreover, a linear association was observed between baseline hemoglobin levels and 3-month poor prognosis after ischemic stroke onset. These results suggested that hemoglobin might be a potential biomarker to predict clinical outcomes at 3 months after ischemic stroke.

The findings of present study may have several important clinical implications. The latest guidelines for the early management of acute ischemic stroke patients from American Heart Association/American Stroke Association (AHA/ASA) have mentioned several

Table 2
Odds ratio and 95% confidence interval of clinical outcomes for quartile of hemoglobin at baseline.

	Hemoglobin, g/L				P_{trend}
	Q1 (< 131)	Q2 (131–142)	Q3 (142–152)	Q4 (≥152)	
Primary outcome: death or major disability (mRS 3–6)					
Model 1	ref	0.93 (0.74–1.16)	1.01 (0.79–1.28)	1.34 (1.04–1.74)	0.021
Model 2	ref	1.00 (0.77–1.29)	1.01 (0.76–1.33)	1.38 (1.03–1.86)	0.043
Major disability (mRS 3–5)					
Model 1	ref	0.98 (0.78–1.25)	1.15 (0.90–1.48)	1.44 (1.11–1.89)	0.004
Model 2	ref	1.07 (0.83–1.39)	1.20 (0.92–1.58)	1.49 (1.11–1.99)	0.006
Death					
Model 1	ref	0.75 (0.45–1.26)	0.44 (0.23–0.84)	0.82 (0.44–1.51)	0.225
Model 2	ref	0.85 (0.49–1.49)	0.43 (0.21–0.85)	0.79 (0.41–1.52)	0.190

Model1, adjusted for age, sex, time from onset to hospitalization, antihypertensive treatment, current smoking, alcohol consumption, body mass index, dyslipidemia, fasting plasma glucose and SBP at baseline, ischemic stroke subtypes, history of hypertension, diabetes and coronary heart disease, and family history of stroke; Model 2, model 1 plus baseline NIHSS score.

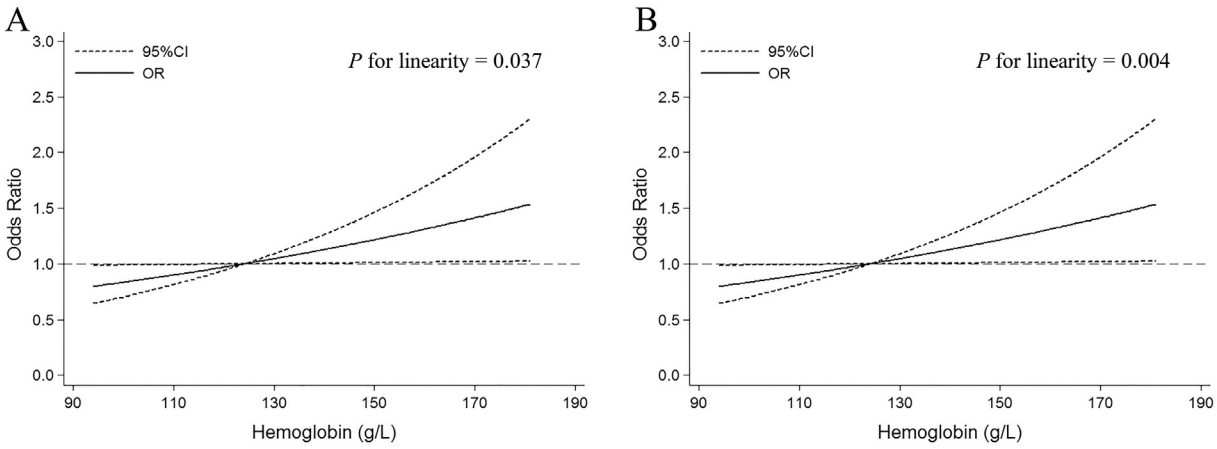

Fig. 2. Linear test of the association between baseline blood hemoglobin and 3-month clinical outcomes.
Odds ratio and 95% confidence interval derived from restricted cubic spline regression, with knots placed at the 5th, 35th, 65th, and 95th percentiles of hemoglobin. Panels adjusted for the same variables as model 2 in Table 2. (A) Primary outcome; (B) Major disability. There was a linear association between hemoglobin levels and 3-month poor prognosis of ischemic stroke (primary outcome: P for linearity = 0.037; major disability: P for linearity = 0.004).

recommendations of general supportive care and emergency treatment, which includes supplemental oxygen, volume expansion/hemodilution and anticoagulants [21]. High levels of hemoglobin are known to increase blood viscosity, which may in turn increase BP and worsen cardiovascular function [22,23]. In addition, it has been suggested that hemoglobin concentrations are significantly higher in individuals with hypertension than in those without hypertension [9,22,24–26]. In the present study, high hemoglobin levels at baseline were associated with poor prognosis of ischemic stroke after adjusting for several important confounders, indicating that hemoglobin can provide additional prognostic information based on the established prognostic factors for ischemic stroke patients.

As a routine laboratory index in clinical practice, it is simple and easy to test hemoglobin levels at acute phase. Therefore, our findings suggest that hemoglobin levels should be evaluated at admission, and patients with high hemoglobin level should be aggressively monitored although the guidelines have not yet recommended the ways to deal with elevated hemoglobin.

In fact, high hemoglobin levels have been reported to be positively associated with the risk of various cardiovascular diseases, including hypertension, atherosclerosis and ischemic stroke [5,9,12,25]. However, reports regarding relationship between hemoglobin levels and prognosis of ischemic stroke are inconsistent so far. Kellert et al. [27] reported that low hemoglobin was associated with poor clinical outcome among 236 ischemic stroke patients. Barlas et al. [14] reported a U-shaped curve relationship between hemoglobin and mortality after stroke among 6951 patients from a hospital-based register database, in which extreme low and extreme high hemoglobin levels were both associated with increased risk of mortality after ischemic stroke. However, the participants in that study is heterogeneous because ischemic stroke patients with thrombolytic therapy and those without thrombolytic therapy are all included. Our study is a multicenter prospective study with a relatively large sample size of acute ischemic stroke patients with elevated systolic BP. In our study, we were not able to analyze the relationship between extreme low hemoglobin and poor prognosis after ischemic stroke because of rare patients with extreme low hemoglobin. However, such characteristics that all participants had elevated systolic BP might enable us to provide more valid appraisal of relationship between high hemoglobin and poor prognosis after ischemic stroke. The present study had rigid quality control procedures for baseline data collection and outcome assessment. Furthermore, comprehensive information about potential confounders were collected and adjusted in the multivariate models. In addition, the participants treated with intravenous thrombolytic therapy which might increase hemoglobin concentrations [28] were excluded, so the hemoglobin levels at baseline were not affected by thrombolytic therapy. We found that elevated hemoglobin was associated with poor outcomes at 3 months among 3883 ischemic stroke patients with an elevated systolic BP. Moreover, elevated hemoglobin remained independently associated with poor prognosis even after adjusting for baseline NIHSS score. We also found that the prognostic value of hemoglobin seems to be stronger for major disability than for death, indicating that elevated hemoglobin

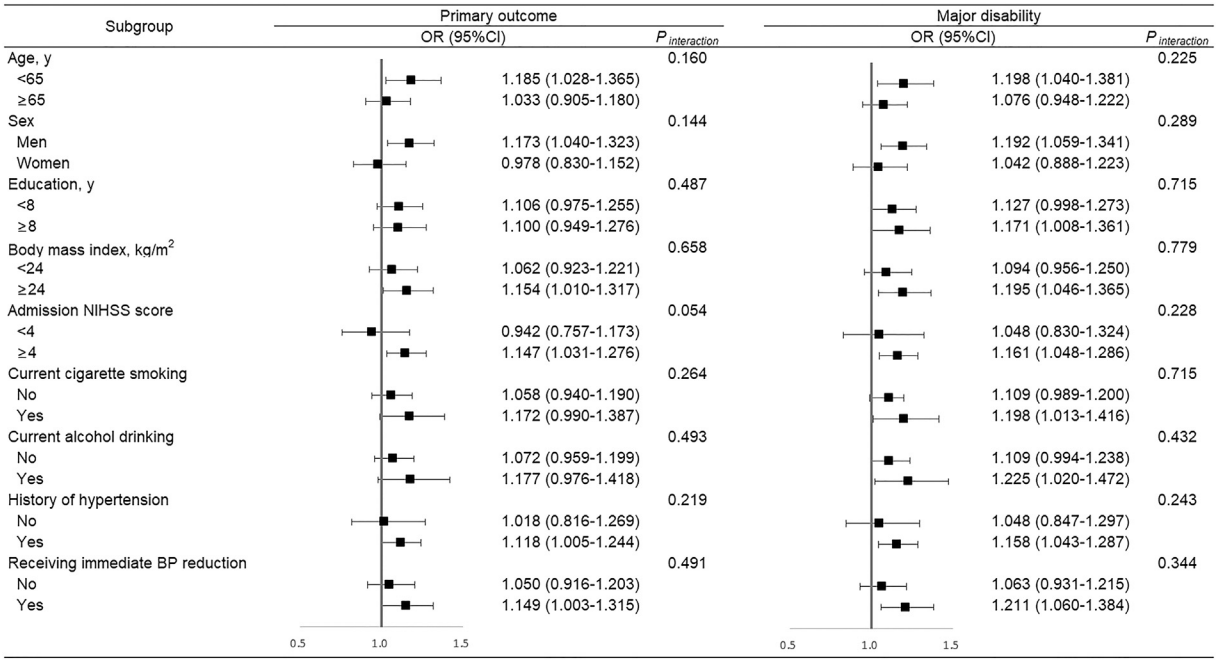

Fig. 3. Subgroup analyses of the association between baseline blood hemoglobin and clinical outcomes of ischemic stroke at 3 months.
In the multivariate models, confounding factors such as age, sex, time from onset to hospitalization, antihypertensive treatment, current smoking, alcohol consumption, body mass index, dyslipidemia, fasting plasma glucose, systolic BP and NIHSS score at baseline, ischemic stroke subtypes, history of hypertension, history of diabetes, history of coronary heart disease and family history of stroke were included unless the variable was used as a subgroup variable.

may mainly affect functional recovery of ischemic stroke patients. In addition, rates of slight stroke in CATIS is relatively high [median NIHSS score: 4 (2–7)]. Therefore, relatively few numbers of death were observed during 3-month follow-up, which might limit our power to detect significant association between hemoglobin and death. Further long-term follow-up studies are required to examine the association between them.

Although the mechanisms underlying association between increased hemoglobin and poor prognosis after ischemic stroke are not clear yet, several potential pathophysiological pathways have been suggested. Elevated BP is common in the acute phase of ischemic stroke [29], which can lead to the activation of the renin-angiotensin-aldosterone system and then result in angiotensin-2 producing, vasoconstriction and erythropoietin production [30]. In addition, endothelial cell damage may increase not only BP but also growth factor level [31],which enhances hematopoiesis and then increases hemoglobin level [32].In addition, both increased blood viscosity [33] and iron overload [34] due to high hemoglobin can affect coronary, cerebral, as well as peripheral blood flow and perfusion [35–37]. Blood rheology is important in the brain microcirculation, and even a small reduction in blood flow may have a huge impact on cerebral function, especially in acute phase of ischemic stroke [38]. Elevated hemoglobin can increase blood viscosity, and further increase peripheral resistance and diminished cardiac output [5]. High hemoglobin levels may also aggregate erythrocyte, and then lead to platelet aggregation and adhesion on the arterial wall [33,37,39].

Our study has some limitations. First, this study was not applicable to assess the association of low hemoglobin levels with poor outcomes after ischemic stroke because of a low rate of anemia among the participants. Second, the selection bias may exist in the present study, but the baseline characteristics of participants are similar to those from the China National Stroke Registry, suggesting that the selection bias may be minimal [40]. Third, we did not collect the data of other high hemoglobin-related factors such as iron intake, the altitude of residence, and lung function/disease, so there may be a possibility of residual confounding in this study.

5. Conclusions

In summary, elevated hemoglobin levels in the acute phase were associated with poor prognosis at 3 months after ischemic stroke. Further prospective studies from other samples of ischemic stroke patients are needed to validate our findings.

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Disclosures

None.

Author contributions

Yonghong Zhang and Jiang He conceived and designed the study. Yanbo Peng, Tan Xu, and Yonghong Zhang coordinated the study. Daoxia Guo, Zhengbao Zhu, Chongke Zhong, Hao Peng, Tian Xu, Aili Wang, Yanbo Peng, Tan Xu, Chung-Shiuan Chen, Yongqiu Li, Zhong Ju, Jing Chen, Yonghong Zhang, and Jiang He oversaw subject recruitment and monitored gathering of clinical data. Daoxia Guo and Zhengbao Zhu conducted the statistical analysis and prepared the paper. Yonghong Zhang and Jiang He revised the paper.

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