MOYAMOYA disease (MMD) is a chronic progressive cerebrovascular disease of unknown etiology. This disease is usually characterized by bilateral internal carotid artery stenosis or occlusion, accompanied by the formation of abnormal vascular network.1,2

Revascularization surgery for symptomatic MMD is considered the standard treatment for preventing further stroke. Encephaloduroarteriosynangiosis (EDAS) is one of the most commonly used indirect vascular reconstruction methods.3–7 However, we found that about 20% of patients had poor postoperative collateral circulation in our previous long-term follow-up studies.8,9 A recent study by our center showed that the formation of collateral vessels in EDAS is primarily driven by angiogenesis, and that the endothelial progenitor cell (EPC) count may be the most critical factor for promoting collateral circulation.10

Studies have shown that statins can effectively reduce the incidence of death and ischemic cardiovascular events in patients with coronary heart disease by reducing cholesterol.11–13 Further animal experiments have shown that statins can also affect the mobilization, proliferation, chemotaxis, and apoptosis of EPCs.14 Importantly, they may suggest that the mobilization of EPCs by statins might represent a useful strategy for clinical therapy of MMD after EDAS. Therefore, we designed this prospective study to confirm the role of atorvastatin in collateral circulation formation induced by EDAS in patients with MMD.

OBJECTIVE This prospective study was designed to confirm the role of atorvastatin in collateral circulation formation induced by encephaloduroarteriosynangiosis (EDAS) in patients with moyamoya disease (MMD).

METHODS Patients who were diagnosed with MMD at the Department of Neurosurgery in the Fifth Medical Center of Chinese PLA General Hospital, Beijing, China, between June 2017 and May 2018 were included. Blood samples were obtained from an antecubital vein and were analyzed using flow cytometry. Endothelial progenitor cells (EPCs) were defined as CD34+CD133+CD45dimKDR+. All patients included in the study underwent EDAS. Patients voluntarily chose whether to undergo atorvastatin treatment after EDAS. The correlation between atorvastatin and good postoperative collateral circulation was evaluated.

RESULTS A total of 106 patients with MMD were included in this study. Fifty-three patients (50%) received atorvastatin treatment. The baseline characteristics did not display statistically significant differences between the atorvastatin-treated and non-atorvastatin groups. Seventy-eight (42.9%) of the 182 hemispheres investigated postoperatively were classified as grade A collateral circulation, 47 (25.8%) as grade B, and 57 (31.3%) as grade C. Multivariate analysis revealed that only atorvastatin was significantly correlated with good collateral circulation after EDAS (p = 0.041).

CONCLUSIONS The results of this prospective clinical trial have indicated that atorvastatin administered at 20 mg daily is safe and effective for the formation of postoperative collateral induced by EDAS.

https://thejns.org/doi/abs/10.3171/2021.6.FOCUS21112

KEYWORDS moyamoya; encephaloduroarteriosynangiosis; atorvastatin
Methods

Patient Selection

Patients who were diagnosed with MMD at the Department of Neurosurgery in the Fifth Medical Center of Chinese PLA General Hospital, Beijing, China, between June 2017 and May 2018 were included in this study (clinical trial registration no. NCT03613701 [clinicaltrials.gov]). The inclusion and exclusion criteria of MMD diagnosis are referred to in previous literature.15–17 All patients were aged ≥ 18 years. All patients were evaluated for liver and kidney function before being included in the study, and patients with abnormal liver or kidney function were excluded.

Study Design

A flow diagram of the study is shown in Fig. 1. A blood sample from each patient was collected before revascularization. All peripheral blood samples were analyzed by flow cytometry.18 EPCs were defined as CD34briCD133+CD45dimKDR+. The EPC count was reported as a percentage of the peripheral blood mononuclear cells (PBMCs). All patients with MMD included in the study underwent EDAS. The specific surgical methods were referred to in previous reports.19

After surgery, the researchers fully explained the intention of this trial and the possible adverse reactions of atorvastatin treatment to all patients. Patients voluntarily chose whether to take atorvastatin; patients who declined to take atorvastatin were included in the control group. Atorvastatin (Pfizer) was administered at 20 mg per night for 8 weeks.20–22 Laboratory tests of liver and kidney function were performed after 8 weeks of medication. Any adverse reactions that occurred were recorded by the follow-up doctor.

DSA images were obtained 6 months after EDAS. The development of collateral circulation of the middle cerebral artery (MCA) through bypass was graded according to the system described by Matsushima et al.23

Clinical Data

The clinical data obtained in this study included sex, age, initial symptoms, hypertension, diabetes mellitus, preoperative angiographic stage, unilateral disease, and posterior circulation involvement (PCI). The angiographic stage was evaluated according to the Suzuki classification.2 The angiographic collateral grade was evaluated according to the system described in our recent study.24 The grading score was based on the collateral circulation and

FIG. 1. Flow diagram of this study.
TABLE 1. Baseline characteristics of patients with MMD in the study

| Variable                  | Value          |
|---------------------------|----------------|
| Mean age, yrs             | 40.5 ± 9.8     |
| Female sex                | 62 (58.5)      |
| Initial symptoms          |                |
| TIA                       | 37 (34.9)      |
| Infarction                | 50 (47.2)      |
| Hemorrhage                | 19 (17.9)      |
| History of risk factors   |                |
| Hypertension              | 16 (15.1)      |
| Diabetes mellitus         | 12 (11.3)      |
| Smoking or drinking       | 19 (17.9)      |
| Unilateral lesions        | 30 (28.3)      |
| Atorvastatin use          | 53 (50.0)      |
| Mean EPCs before EDAS     | 0.061% ± 0.053%|

Values represent the number of patients (%) or mean ± SD.

TABLE 2. Comparison of baseline characteristics and angiographic outcome between the atorvastatin and control groups

| Variable                  | Value          |
|---------------------------|----------------|
| Age at symptom onset      | 40.5 ± 9.8     |
| Sex                       |                |
| Male                      |                |
| Female                    |                |
| Initial symptoms          |                |
| TIA                       | 37 (34.9)      |
| Infarction                | 50 (47.2)      |
| Hemorrhage                | 19 (17.9)      |
| History of risk factors   |                |
| Hypertension              | 16 (15.1)      |
| Diabetes mellitus         | 12 (11.3)      |
| Smoking or drinking       | 19 (17.9)      |
| Unilateral lesions        | 30 (28.3)      |
| Atorvastatin use          | 53 (50.0)      |
| Mean EPCs before EDAS     | 0.061% ± 0.053%|

Values represent the number of patients (%) or mean ± SD.

Association of Postoperative Collateral Formation With Atorvastatin

Because of the unilateral lesion in 30 patients, a total of 182 hemispheres were treated with EDAS. Cerebral arteriography was performed 6 months after EDAS to assess the efficacy of synangiosis and to guide subsequent disease management. Seventy-eight (42.9%) of the 182 hemispheres investigated were classified as grade A collateral circulation, 47 (25.8%) as grade B, and 57 (31.3%) as grade C.

Results

Patient Characteristics

A total of 106 patients with MMD were included in this study. The clinical characteristics of the patients are summarized in Table 1. The mean age at symptom onset was 40.5 ± SD 9.8 years, and there were 62 female and 44 male patients. Among the patients with MMD, a history of hypertension was reported in 16 patients (15.1%), a history of diabetes in 12 (11.3%), and previous tobacco or alcohol use in 19 (17.9%). As the initial symptoms, 37 patients (34.9%) exhibited transient ischemic attacks, 50 patients (47.2%) exhibited infarction symptoms, and 19 patients (17.9%) exhibited hemorrhagic symptoms. Thirty patients exhibited unilateral disease. The EPC count in the patients with MMD was 0.061% ± 0.053% (expressed as the percentage of PBMCs). According to study design, 53 (50%) patients received atorvastatin treatment voluntarily.

Table 2 shows the comparison of baseline characteristics and angiographic outcome between the atorvastatin and control groups. Baseline characteristics included age at symptom onset, sex, history of risk factors, EPC count before EDAS, unilateral lesions, symptoms in hemispheres, PCI, Suzuki stage, TTP delay, and collateral grade; none displayed statistically significant differences between the two groups (p > 0.05).

Statistical Analysis

The chi-square test was used to analyze categorical variables, and the independent Student t-test or ANOVA was used to compare continuous variables. The Mann-Whitney U-test or the Kruskal-Wallis test was performed on variables that did not follow a normal distribution. Univariate analysis was used to assess the correlation between age, sex, initial symptoms at diagnosis, collateral grade, PCI, atorvastatin use, and the EPC count and collateral circulation after EDAS. The variables with significant association (p < 0.05) were selected and subjected to multiple linear regression analysis. A p value < 0.05 was considered statistically significant.

Results

Patient Characteristics

A total of 106 patients with MMD were included in this study. The clinical characteristics of the patients are summarized in Table 1. The mean age at symptom onset was 40.5 ± SD 9.8 years, and there were 62 female and 44 male patients. Among the patients with MMD, a history of hypertension was reported in 16 patients (15.1%), a history of diabetes in 12 (11.3%), and previous tobacco or alcohol use in 19 (17.9%). As the initial symptoms, 37 patients (34.9%) exhibited transient ischemic attacks, 50 patients (47.2%) exhibited infarction symptoms, and 19 patients (17.9%) exhibited hemorrhagic symptoms. Thirty patients exhibited unilateral disease. The EPC count in the patients with MMD was 0.061% ± 0.053% (expressed as the percentage of PBMCs). According to study design, 53 (50%) patients received atorvastatin treatment voluntarily.

Table 2 shows the comparison of baseline characteristics and angiographic outcome between the atorvastatin and control groups. Baseline characteristics included age at symptom onset, sex, history of risk factors, EPC count before EDAS, unilateral lesions, symptoms in hemispheres, PCI, Suzuki stage, TTP delay, and collateral grade; none displayed statistically significant differences between the two groups (p > 0.05).

Association of Postoperative Collateral Formation With Atorvastatin

Because of the unilateral lesion in 30 patients, a total of 182 hemispheres were treated with EDAS. Cerebral arteriography was performed 6 months after EDAS to assess the efficacy of synangiosis and to guide subsequent disease management. Seventy-eight (42.9%) of the 182 hemispheres investigated were classified as grade A collateral circulation, 47 (25.8%) as grade B, and 57 (31.3%) as grade C.

Results of the univariate analysis are summarized in Table 3. Patients who were treated with atorvastatin could get better postoperative collateral circulation (grade A or B) than the control group (p = 0.006). The postoperative angiographic findings also showed that a good Matsushima grade was correlated with infarction symptoms (p = 0.034) and a higher EPC count before EDAS (p > 0.99). The variables with significant correlation were subjected to multiple linear regression analysis. The multivariate analysis revealed that only atorvastatin was significantly correlated with good collateral circulation after EDAS (p = 0.041).

Figures 2 and 3 show representative patients from each of the two groups. A 50-year-old female whose initial symptom was cerebral infarction had an EPC percentage in peripheral blood of 0.039% before EDAS, which was far below the average level. Atorvastatin was administered regularly after the operation. Excellent collateral circulation was obtained in both hemispheres after 6 months (Fig. 2). In contrast, Fig. 3 shows a 43-year-old...
TABLE 2. Comparison of baseline characteristics and angiographic outcome between the atorvastatin and control groups

| Variable                  | Atorvastatin Group (n = 53) | Control Group (n = 53) | p Value |
|---------------------------|-----------------------------|------------------------|---------|
| Mean age, yrs             | 42.4 ± 9.9                  | 38.6 ± 9.3             | 0.551   |
| Female sex                | 25 (47.2)                   | 37 (69.8)              | 0.073   |
| History of risk factors   | 23 (43.4)                   | 24 (45.3)              | 0.845   |
| Mean EPCs before EDAS     | 0.066 ± 0.056%              | 0.055 ± 0.043%         | 0.183   |
| Unilateral lesion         | 15 (28.3)                   | 15 (28.3)              | >0.99   |
| Symptoms in hemispheres   |                             |                        | 0.254   |
| Asymptomatic              | 34 (37.3)                   | 31 (34.1)              |         |
| TIA                       | 18 (19.8)                   | 25 (27.5)              |         |
| Infarction                | 32 (35.2)                   | 23 (25.3)              |         |
| Hemorrhage                | 7 (7.7)                     | 12 (13.2)              |         |
| PCI                       | 19 (20.9)                   | 23 (25.3)              | 0.482   |
| Suzuki stage              |                             |                        | 0.805   |
| I                         | 9 (9.9)                     | 8 (8.8)                |         |
| II                        | 18 (19.8)                   | 13 (14.3)              |         |
| III                       | 18 (19.8)                   | 19 (20.9)              |         |
| IV                        | 19 (20.9)                   | 22 (24.2)              |         |
| V                         | 15 (16.5)                   | 12 (13.2)              |         |
| VI                        | 12 (13.2)                   | 17 (18.7)              |         |
| Collateral grade          |                             |                        | 0.797   |
| Poor (stage I)            | 35 (38.5)                   | 37 (40.7)              |         |
| Fair (stage II)           | 30 (33.0)                   | 32 (35.2)              |         |
| Good (stage III)          | 26 (28.6)                   | 22 (24.2)              |         |
| Mean TTP delay, secs      | Before EDAS 4.55 ± 2.18     | 3.78 ± 1.98            | 0.644   |
| 6 mos after EDAS          | 3.41 ± 1.67                 | 3.01 ± 2.41            | 0.481   |

1Values represent the number of patients (%) or mean ± SD unless indicated otherwise.

Discussion

Atorvastatin, as a classic lipid-lowering drug, has been widely used in the cardiovascular field. Moreover, some reports have confirmed that atorvastatin could also promote angiogenesis in models of stroke and brain injury.25–27 However, to the best of our knowledge, no studies have reported the use of atorvastatin in the treatment of MMD, and our study may provide a new idea for drug-assisted therapy after indirect revascularization.

In this prospective study, we found that the extent of postoperative collateral formation in patients with atorvastatin treatment was significantly higher than the control group, which means that atorvastatin could help patients have a better surgical outcome. This is of great significance for MMD patients, especially for those with severe cerebral ischemia. Patients can take atorvastatin to further strengthen the formation of postoperative collateral circulation, so as to better prevent the occurrence of stroke in the future.

At present, research on the effect of statins on EPCs in human peripheral blood has been mostly concentrated in the cardiovascular field. It has been confirmed that statins can affect the mobilization, proliferation, chemotaxis, and apoptosis of EPCs.28–30 Vasa et al.31 treated patients with stable coronary heart disease confirmed by arteriography with atorvastatin. The authors found that the number of circulating EPCs increased significantly, and that the migration ability of EPCs isolated and cultured from peripheral blood significantly enhanced. Schöming et al.32 observed in patients with acute myocardial infarction that the number of circulating EPCs in the peripheral blood was positively correlated with statins. Landmesser et al.33 also observed in patients with chronic heart failure that after 4 weeks of treatment with simvastatin 10 mg/day, the number of EPCs isolated and cultured from peripheral blood increased significantly. The aforementioned studies have shown that statins can significantly promote the number and function of EPCs; however, the specific mechanism of atorvastatin that promotes angiogenesis is not yet clear. Based on the results in previous studies, we speculate that statin-induced stimulation of the Akt/endothelial nitric oxide synthase pathway might contribute to the observed effects of statins on the functional improvement of EPCs.30–34

It is undeniable that 33.3% of the hemispheres in our study did not achieve good collateral circulation after atorvastatin treatment. The process of angiogenesis is affected by many factors, and EPCs are only one of the important components. In future efforts, we will carry out a multicenter, large-sample, prospective randomized controlled study to further clarify the role of atorvastatin in patients with MMD after surgery. The specific molecular biology mechanism should also be explored at the same time.

Limitations

This study has several limitations. First, it is not a prospective randomized controlled study. Patients voluntarily

Drug Safety and Follow-Up

According to the follow-up laboratory tests, 8 patients (15.1%) presented with mild liver abnormalities and 2 patients (3.8%) with mild kidney abnormalities, but none required treatment. No patient experienced a severe adverse drug reaction or event, which included the following: death, life-threatening or permanent or significant disability, permanent functional injury to the organs, hospitalization for emergencies, or prolonged hospitalization. During the follow-up period after EDAS, no patient had cerebral infarction or cerebral hemorrhage caused by MMD.

male in the control group who also had cerebral infarction as the first symptom and had an EPC percentage of 0.027% before EDAS. Reexamination of cerebral angiography 6 months later indicated poor collateral circulation formation.

Support and Funding

This study was supported by grants from the National Natural Science Foundation of China (81671192) and Jiangsu Medical Science and Technology Development Foundation (BL2019026).
chose whether to undergo atorvastatin treatment or not, which may cause statistical bias on the results of the experiment. Second, our study was conducted at a single center, the sample size was small, and it included a patient population that lacked heterogeneity. Third, due to geographic or economic reasons, some patients underwent cerebral angiography review in other hospitals. Therefore, we could not detect the number of EPCs in peripheral

| Variable                          | Grade A (n = 78) | Grade B (n = 47) | Grade C (n = 57) | p Value |
|-----------------------------------|-----------------|-----------------|-----------------|---------|
| Mean age, yrs                     | 41.1 ± 9.8      | 38.7 ± 9.7      | 41.2 ± 9.8      | 0.997   |
| Symptoms in hemispheres           |                 |                 |                 | 0.034   |
| Asymptomatic                      | 26 (33.3)       | 16 (34.0)       | 23 (40.4)       |         |
| TIA                               | 21 (26.9)       | 11 (23.4)       | 11 (19.3)       |         |
| Infarction                        | 27 (34.6)       | 17 (36.2)       | 11 (19.3)       |         |
| Hemorrhage                        | 4 (5.1)         | 3 (6.4)         | 12 (21.1)       |         |
| PCI                               | 18 (23.1)       | 13 (27.7)       | 11 (19.3)       | 0.602   |
| Collateral grade                  |                 |                 |                 | 0.194   |
| Poor (stage I)                    | 35 (44.9)       | 20 (42.6)       | 17 (29.8)       |         |
| Fair (stage II)                   | 28 (35.9)       | 15 (31.9)       | 19 (33.3)       |         |
| Good (stage III)                  | 15 (19.2)       | 12 (25.5)       | 21 (36.8)       |         |
| Atorvastatin use                  |                 |                 |                 | 0.006   |
| Yes                               | 42 (53.8)       | 30 (63.8)       | 19 (33.3)       |         |
| No                                | 36 (46.2)       | 17 (36.2)       | 38 (66.7)       |         |
| Mean EPCs before EDAS             | 0.082 ± 0.063%  | 0.064 ± 0.033%  | 0.043 ± 0.035%  | <0.001  |

Values represent the number of patients (%) or mean ± SD unless indicated otherwise.

FIG. 2. A patient from the atorvastatin group. A: Flow cytometry analysis of a 50-year-old female who had cerebral infarction as the first symptom and an EPC percentage in the peripheral blood measured before EDAS of 0.039%, which was far below the average level. B and C: Bilateral internal carotid angiograms, lateral views, before EDAS. Atorvastatin was administered regularly after the surgery. D and E: Excellent collateral circulation induced by EDAS (Matsushima grade A) was obtained in both hemispheres after 6 months. -A = area; APC = allophycocyanin; Comp = compensation; FITC = fluorescein isothiocyanate; FSC = forward scatter; PE = phycoerythrin; PerCP = peridinin chlorophyll protein complex; SSC = side scatter.
blood 6 months after surgery in all study patients. In addition, the molecular mechanism of atorvastatin affecting the formation of surgical collateral circulation still needs to be further explored.

Conclusions
This prospective clinical trial finds that atorvastatin administered at 20 mg daily is safe and effective for the formation of postoperative collateral induced by EDAS. Patients with severe ischemic symptoms are more likely to obtain a better surgical outcome.

Acknowledgments
This study was supported by a grant from the National Natural Science Foundation of China (grant no. 81571136).

Reference
1. Choi JU, Kim DS, Kim EY, Lee KC. Natural history of moyamoya disease: comparison of activity of daily living in surgery and non surgery groups. Clin Neurol Neurosurg. 1997;99(suppl 2):S11-S18.
2. Suzuki J, Takaku A. Cerebrovascular “moyamoya” disease. Disease showing abnormal net-like vessels in base of brain. Arch Neurol. 1969;20(3):288-299.
3. Houkin K, Ishikawa T, Yoshimoto T, Abe H. Direct and indirect revascularization for moyamoya disease surgical techniques and peri-operative complications. Clin Neurol Neurosurg. 1997;99(suppl 2):S142-S145.
4. Ishikawa T, Houkin K, Kamiyama H, Abe H. Effects of surgical revascularization on outcome of patients with pediatric moyamoya disease. Stroke. 1997;28(6):1170-1173.
5. Kim SK, Cho BK, Phi JH, Lee JY, Chae JH, Kim KJ, et al. Pediatric moyamoya disease: an analysis of 410 consecutive cases. Ann Neurol. 2010;68(1):92-101.
6. Matsushima Y, Aoyagi M, Suzuki R, Tabata H, Ohno K. Perioperative complications of encephalo-duro-arterio-synangiosis: prevention and treatment. Surg Neurol. 1991;36(5):343-353.
7. Scott RM, Smith JL, Robertson RL, Madsen JR, Soriano SG, Rockoff MA. Long-term outcome in children with moyamoya syndrome after cranial revascularization by pial synangiosis. J Neurosurg. 2004;100(2)(Suppl Pediatrics):142-149.
8. Zhang Y, Bao XY, Duan L, Yang WZ, Li DS, Zhang ZS, et al. Encephaloduroarteriosynangiosis for pediatric moyamoya disease: long-term follow-up of 100 cases at a single center. J Neurosurg Pediatr. 2018;22(2):173-180.
9. Bao XY, Zhang Y, Wang QN, Zhang Q, Wang H, Zhang ZS, et al. Long-term outcomes after encephaloduroarteriosynangiosis in adult patients with moyamoya disease presenting with ischemia. World Neurosurg. 2018;115:e482-e489.
10. Wang QN, Zou ZX, Wang XP, Zhang Q, Zhao YQ, Duan L, Bao XY. Endothelial progenitor cells induce angiogenesis: a potential mechanism underlying neovascularization in encephaloduroarteriosynangiosis. Transl Stroke Res. 2021;12(2):357-365.
11. Scandinavian Simvastatin Survival Study Group. Randomised trial of cholesterol lowering in 4444 patients with coronary heart disease: the Scandinavian Simvastatin Survival Study (4S). Lancet. 1994;344(8934):1383-1389.
12. Long-Term Intervention with Pravastatin in Ischaemic Disease (LIPID) Study Group. Prevention of cardiovascular events and death with pravastatin in patients with coronary
heart disease and a broad range of initial cholesterol levels. N Engl J Med. 1998;339(19):1349-1357.
13. Sacks FM, Pfeffer MA, Moye LA, Rouleau JL, Rutherford JD, Cole TG, et al. The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol levels. Cholesterol and Recurrent Events Trial investigators. N Engl J Med. 1996;335(14):1001-1009.
14. Vasa M, Fichtlscherer S, Adler K, Aicher A, Martin H, Zeiher AM, Dimmeler S. Increase in circulating endothelial progenitor cells by statin therapy in patients with stable coronary artery disease. Circulation. 2001;103(24):2885-2890.
15. Research Committee on the Pathology and Treatment of Spontaneous Occlusion of the Circle of Willis. Guidelines for diagnosis and treatment of moyamoya disease (spontaneous occlusion of the circle of Willis). Neurol Med Chir (Tokyo). 2012;52(5):245-266.
16. Bang OY, Fujimura M, Kim SK. The pathophysiology of moyamoya disease: an update. J Stroke. 2016;18(1):12-20.
17. Fujimura M, Tominaga T. Diagnosis of moyamoya disease: international standard and regional differences. Neurol Med Chir (Tokyo). 2015;55(3):189-193.
18. Duda DG, Cohen KS, Scadden DT, Jain RK. A protocol for phenotypic detection and enumeration of circulating endothelial cells and circulating progenitor cells in human blood. Nat Protoc. 2007;2(4):805-810.
19. Hung CC, Tu YK, Su CF, Lin LS, Shih CJ. Epidemiological study of moyamoya disease in Taiwan. Clin Neurol Neurosurg. 1997;99(suppl 2):S23-S25.
20. Bakker-Arkema RG, Davidson MH, Goldstein RJ, Davignon J, Isaacsohn JL, Weiss SR, et al. Efficacy and safety of a new HMG-CoA reductase inhibitor, atorvastatin, in patients with hypertriglyceridemia. JAMA. 1998;279(7):1349-1357.
21. Kureishi Y, Luo Z, Shiojima I, Bialik A, Fulton D, Lefer DJ, et al. The HMG-CoA reductase inhibitor simvastatin activates the protein kinase Akt and promotes angiogenesis in normocholesterolemic animals. Nat Med. 2000;6(9):1004-1010.
22. Dimmeler S, Fleming I, Fisslthaler B, Herrmann C, Busse R, Zeiher AM. Activation of nitric oxide synthase in endothelial cells by Akt-dependent phosphorylation. Nature. 1999;399(6736):601-605.
23. Matsushima T, Inoue T, Suzuki SO, Fujii K, Fukui M, Hasuo K. Surgical treatment of moyamoya disease in pediatric patients—comparison between the results of indirect and direct revascularization procedures. Neurosurgery. 1992;31(3):401-405.
24. Liu ZW, Han C, Zhao F, Qiao PG, Wang H, Bao XY, et al. Collateral circulation in moyamoya disease: a new grading system. Stroke. 2019;50(10):2708-2715.
25. Potey C, Ouk T, Petrault O, Petrault M, Berezowski V, Salleron J, et al. Early treatment with atorvastatin exerts parenchymal and vascular protective effects in experimental cerebral ischaemia. Br J Pharmacol. 2015;172(21):5188-5198.
26. Rodríguez-PereA AL, Gutierrez-Vargas J, Cardona-Gómez GP, Guarin CJ, Rojas M, Hernández PA. Atorvastatin modulates regulatory T cells and attenuates cerebral damage in a model of transient middle cerebral artery occlusion in rats. J Neuroimmune Pharmacol. 2017;12(1):152-162.
27. Chen J, Zacharek A, Li A, Cui X, Roberts C, Lu M, Chopp M. Atorvastatin promotes presenilin-1 expression and Notch1 activity and increases neural progenitor cell proliferation after stroke. Stroke. 2008;39(1):220-226.
28. Schömig K, Busch G, Steppich B, Sepp D, Kaufmann J, Stein A, et al. Interleukin-8 is associated with circulating CD133+ progenitor cells in acute myocardial infarction. Eur Heart J. 2006;27(9):1032-1037.
29. Landmesser U, Buhlmann F, Mueller M, Spiekermann S, Kirchoff N, Schulz S, et al. Simvastatin versus ezetimibe: pleiotropic and lipid-lowering effects on endothelial function in humans. Circulation. 2005;111(18):2356-2363.
30. Laufs U, Liao JK. Post-transcriptional regulation of endothelial nitric oxide synthase mRNA stability by Rho GTPase. J Biol Chem. 1998;273(37):24266-24271.
31. Laufs U, Liao JK. Post-transcriptional regulation of endothelial nitric oxide synthase mRNA stability by Rho GTPase. J Biol Chem. 1998;273(37):24266-24271.
32. Dimmeler S, Finger C, Fisslthaler B, Herrmann C, Busse R, Zeiher AM. Activation of nitric oxide synthase in endothelial cells by Akt-dependent phosphorylation. Nature. 1999;399(6736):601-605.
33. Dimmeler S, Dernbach E, Zeiher AM. Phosphorylation of the endothelial nitric oxide synthase at ser-1177 is required for VEGF-induced endothelial cell migration. FEBS Lett. 2000;477(3):258-262.
34. Dimmeler S, Zeiher AM. Akt takes center stage in angiogenesis signaling. Circ Res. 2000;86(1):4-5.

Disclosures
The authors report no conflict of interest concerning the materials or methods used in this study or the findings specified in this paper.

Author Contributions
Conception and design: Duan, Zhao. Acquisition of data: QN Wang, Bao, XP Wang, Zhang. Drafting the article: QN Wang, Li. Critically revising the article: Zou. Statistical analysis: Zou, Li.

Correspondence
Lian Duan: Chinese PLA General Hospital, Beijing, China. duanlian307@sina.com.