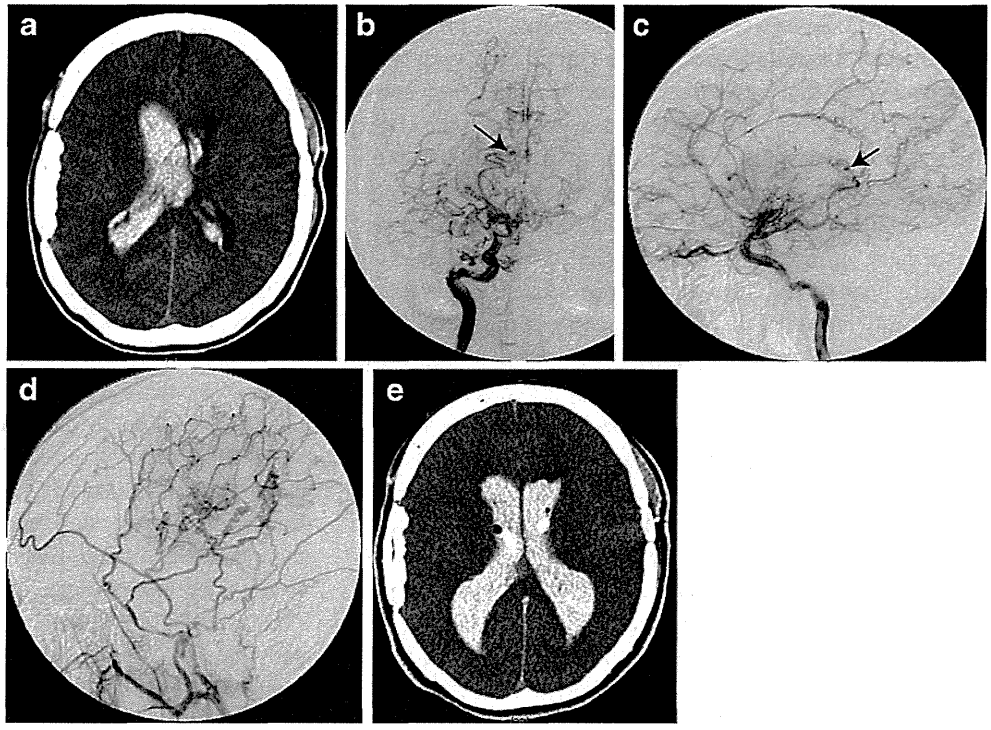

Fig. 4 Case 4. **a** Computed tomography at first hemorrhage; **b–d** angiography after the first hemorrhage; anteroposterior (**b**) and lateral (**c**) views of the internal carotid angiography after first hemorrhage revealing occlusion of the internal carotid artery and dilated posterior choroidal artery accompanied with microaneurysms (*arrows*); **d** external carotid angiography showing good patency of the bypass; **e** computed tomography at second hemorrhage demonstrating massive intraventricular hemorrhage, resulting in fatal outcome

Considering that most children had experienced an ischemic symptom at disease onset, the incidence of ischemic stroke revealed in our study (0.1 % per year) seems quite low. In our series, only one case experienced an ischemic stroke, an event that might not have occurred had he not experienced a severe head injury. Bypass surgery for moyamoya disease is usually successful at improving cerebral blood flow [13, 28, 34]. While the natural history of pediatric moyamoya disease is devastating [2], revascularization surgery can be effective at preventing ischemic stroke for decades.

In the present study, hemorrhage occurred more frequently than ischemic stroke during the follow-up period. This finding is coincident with the previous studies (Table 3) [12, 27]. Interestingly, all late hemorrhages in our study occurred in those patients who had first experienced ischemic symptoms. From a long-term perspective, hemorrhages should receive more attention than strokes do because hemorrhage affects outcome more severely [18, 25, 38]. On the other hand, our results do not coincide with those reported by Scott et al. [31], which showed 4 late-onset ischemic strokes in their 126 cases but no hemorrhage after pial synangiosis without direct

Table 3 Incidence of late cerebrovascular events in previous and present studies

	Mukawa et al.	Imaizumi et al.	Present study
Treatment	Indirect bypass	Indirect and direct bypasses, or conservative[a]	Direct bypass
No. of patients	172	25	58
Mean follow-up period in years (range)	14.3 (3–32)	18.8 (NA)	18.1 (9–33.7)
Follow-up rate	83 %	80.6 %	96.6 %
Total person-years	2,459.6[b]	470.0[b]	986.6
Late cerebrovascular events			
No. and mode	3 strokes, 3 hemorrhages[c]	1 stroke, 3 hemorrhages	1 stroke[d], 3 hemorrhages[e]
Incidence, % per year (95 % CI)	0.24 (0.11–0.54)	0.85 (0.32–2.27)	0.41 (0.15–1.08)

CI confidence interval, *NA* not available

[a] Including 9 cases treated by indirect bypass, 1 by direct bypass, and 15 conservatively

[b] Approximated by multiplying the number of samples by mean follow-up period because the person-years were not readily available

[c] All three patients experienced a second hemorrhage

[d] The stroke occurred after a severe head injury and emergent craniotomy

[e] One of the three patients experienced second hemorrhage

bypasses. The controversy might be attributable to a difference in race, surgical procedure, or target disease; their study was conducted in North America and included moyamoya syndrome comprising autoimmune disease, meningitis, brain tumor, Down syndrome, neurofibromatosis type 1, and so on.

Revascularization surgeries are assumed to reduce the hemodynamic burden on moyamoya vessels and thus to prevent hemorrhage in moyamoya disease [23]. Moyamoya vessels and peripheral artery aneurysms can decrease if bypasses are successfully patent [7, 22]. In some cases in our study, however, the moyamoya vessels or microaneurysms in the posterior circulation remained despite the good patency of bypasses, resulting in late hemorrhage (Cases 2 and 4). One possible explanation for this condition is that the bypass flow might be insufficient to reduce the hemodynamic burden to moyamoya vessels, as the bypass flow might depend more on the severity of preoperative ischemia (indicating greater demand for blood flow) than on the development of moyamoya vessels. Another possible reason is, as Goda et al. [7] reported, that the fragile collateral formation from posterior circulation might not fully decrease after revascularization surgery for anterior circulation. The involvement of a steno-occlusive lesion in posterior circulation thus might be a risk factor for late-onset intracranial hemorrhage. Although additional direct or indirect revascularization to posterior cerebral artery might be effective at improving remaining ischemia [9, 11, 17], whether this treatment option can also reduce the risk of future hemorrhage remains unproved. Determining underlying risk factor and treatment for late hemorrhage are an important issue to be addressed.

In the present study, all late hemorrhages occurred in patients aged in their mid- or late-20s after more than 13 years had passed since bypass surgery. This result is coincident with that reported by Mukawa et al. [27], in which late hemorrhage occurred between 8 and 21 years after revascularization surgery. According to the theory that bleeding in adult patients results from long collateral vessel exposure to hemodynamic burden from a young age, the incidence of hemorrhage may be increasing with the duration of the disease. A follow-up period of less than 10 years might result in underestimation of the incidence of late-onset hemorrhage. This might partially explain the fact that some long-term follow-up studies after surgical revascularization detected no late-onset hemorrhage [4, 7, 21]. In cross-sectional studies, the onset age of hemorrhagic-type moyamoya disease is distributed mainly throughout the 30s and 40s [18, 25, 38]. The mean age of hemorrhage onset in our study seemed younger than that revealed in the cross-sectional studies. This might be attributed to longer disease duration in our study population, which includes only patients with juvenile-onset moyamoya disease. Further follow-up might be important because many patients in our study had not yet reached their 30s or 40s (Table 1).

Because the incidence of initial hemorrhage in the natural course of moyamoya disease remains unclear, our study did not strictly answer the question of whether bypass surgery in childhood is effective at preventing future hemorrhage. However, the incidence of hemorrhage revealed in the present study (0.3 % per year) is far lower than that of recurrent hemorrhage in moyamoya disease (7.09 % per year) [18]. Further investigations might be needed to elucidate whether bypass surgery is effective at primary prevention of hemorrhage in moyamoya disease.

Several reports on indirect bypass have illustrated late cerebrovascular events in pediatric patients with moyamoya disease [12, 27]. To our knowledge, however, no previous study of direct bypass has focused on the incidence of late cerebrovascular events except for a very large study including both pediatric and adult patients [8]. Our result suggests that the incidence of such events after direct bypass is comparable to that after indirect bypass because of the overlap of 95 % CIs (Table 3). This speculation is consistent with a recent review showing that direct and indirect bypasses have equivalent long-term effectiveness [6]. Recent studies have reported excellent results of direct bypass for moyamoya disease [8, 21]. Direct comparisons between direct and indirect bypasses may be needed in future studies to determine whether both surgical options exhibit an equivalent incidence of late onset stroke.

Limitations

Our study included two (3.4 %) patients lost to follow-up. Despite the high follow-up rate compared with previous studies, this loss might induce selection bias if it involves serious outcomes.

Although age and sex distributions in the present study were similar to those in large epidemiological studies of moyamoya disease [1, 20, 37], our use of hospital-based sampling might not accurately reflect the population of pediatric patients with moyamoya disease.

Our results revealed normalized rCBF long after bypass surgery, but the number of patients undergoing SPECT is limited. Further investigation of long-term changes in rCBF and angiographical stages is required to determine whether these factors affect late hemorrhage.

Conclusions

The objective of the present study was to estimate the incidence of late cerebrovascular events after direct bypass surgery in pediatric patients with moyamoya disease. The results suggest that such patients remain at risk of a late cerebrovascular event (0.41 % per year) even after revascularization surgery, although the incidence seems quite low compared with the natural history of moyamoya disease. Among late

cerebrovascular events, hemorrhage might be more common than ischemic stroke and should receive more attention. Late hemorrhage might be more likely to occur in patients in their late 20s who had undergone surgery more than 10 years previously, suggesting the importance of careful long-term follow-up.

Acknowledgments We are grateful to Dr. Takeo Nakayama, Professor at the Department of Health Informatics, Kyoto University School of Public Health, for providing statistical advice. We also thank Yuko Tanaka for her efforts at collecting patient records.

Conflicts of interest None.

References

1. Baba T, Houkin K, Kuroda S (2008) Novel epidemiological features of moyamoya disease. J Neurol Neurosurg Psychiatry 79:900–904
2. Choi JU, Kim DS, Kim EY, Lee KC (1997) Natural history of moyamoya disease: comparison of activity of daily living in surgery and non surgery groups. Clin Neurol Neurosurg 99(Suppl 2):S11–S18
3. Czabanka M, Pena-Tapia P, Scharf J, Schubert GA, Munch E, Horn P, Schmiedek P, Vajkoczy P (2011) Characterization of direct and indirect cerebral revascularization for the treatment of European patients with moyamoya disease. Cerebrovasc Dis 32:361–369
4. Darwish B, Besser M (2005) Long term outcome in children with Moyamoya disease: experience with 16 patients. J Clin Neurosci 12: 873–877
5. Fukui M (1997) Guidelines for the diagnosis and treatment of spontaneous occlusion of the circle of Willis ('moyamoya' disease). Research Committee on Spontaneous Occlusion of the Circle of Willis (Moyamoya Disease) of the Ministry of Health and Welfare, Japan. Clin Neurol Neurosurg 99(2):S238–S240
6. Fung LW, Thompson D, Ganesan V (2005) Revascularisation surgery for paediatric moyamoya: a review of the literature. Childs Nerv Syst 21:358–364
7. Goda M, Isono M, Ishii K, Kamida T, Abe T, Kobayashi H (2004) Long-term effects of indirect bypass surgery on collateral vessel formation in pediatric moyamoya disease. J Neurosurg 100:156–162
8. Guzman R, Lee M, Achrol A, Bell-Stephens T, Kelly M, Do HM, Marks MP, Steinberg GK (2009) Clinical outcome after 450 revascularization procedures for moyamoya disease. Clinical article. J Neurosurg 111:927–935
9. Hayashi T, Shirane R, Tominaga T (2009) Additional surgery for postoperative ischemic symptoms in patients with moyamoya disease: the effectiveness of occipital artery-posterior cerebral artery bypass with an indirect procedure: technical case report. Neurosurgery 64:E195–E196, discussion E196
10. Iida H, Itoh H, Bloomfield PM, Munaka M, Higano S, Murakami M, Inugami A, Eberl S, Aizawa Y, Kanno I et al (1994) A method to quantitate cerebral blood flow using a rotating gamma camera and iodine-123 iodoamphetamine with one blood sampling. Eur J Nucl Med 21:1072–1084
11. Ikeda A, Yamamoto I, Sato O, Morota N, Tsuji T, Seguchi T (1991) Revascularization of the calcarine artery in moyamoya disease: OA-cortical PCA anastomosis—case report. Neurol Med Chir (Tokyo) 31:658–661
12. Imaizumi T, Hayashi K, Saito K, Osawa M, Fukuyama Y (1998) Long-term outcomes of pediatric moyamoya disease monitored to adulthood. Pediatr Neurol 18:321–325
13. Ishii R, Takeuchi S, Ibayashi K, Tanaka R (1984) Intelligence in children with moyamoya disease: evaluation after surgical treatments with special reference to changes in cerebral blood flow. Stroke 15: 873–877
14. Karasawa J, Kikuchi H, Furuse S, Sakaki T, Yoshida Y (1977) A surgical treatment of "moyamoya" disease "encephalo-myo synangiosis". Neurol Med Chir (Tokyo) 17:29–37
15. Karasawa J, Kikuchi H, Kuriyama Y, Sawada T, Kuro M, Kobayashi K, Koike T, Mitsugi T (1981) Cerebral hemodynamics in "moyamoya" disease—II. Measurements of cerebral circulation and metabolism by use of the argon desaturation method in pre- and post-neurosurgical procedures. Neurol Med Chir (Tokyo) 21:1161–1168
16. Karasawa J, Touho H, Ohnishi H, Miyamoto S, Kikuchi H (1992) Long-term follow-up study after extracranial-intracranial bypass surgery for anterior circulation ischemia in childhood moyamoya disease. J Neurosurg 77:84–89
17. Karasawa J, Touho H, Ohnishi H, Miyamoto S, Kikuchi H (1993) Cerebral revascularization using omental transplantation for childhood moyamoya disease. J Neurosurg 79:192–196
18. Kobayashi E, Saeki N, Oishi H, Hirai S, Yamaura A (2000) Long-term natural history of hemorrhagic moyamoya disease in 42 patients. J Neurosurg 93:976–980
19. Kruskal WH, Wallis WA (1952) Use of Ranks in One-Criterion Variance Analysis. J Am Stat Assoc 47:583–621
20. Kuriyama S, Kusaka Y, Fujimura M, Wakai K, Tamakoshi A, Hashimoto S, Tsuji I, Inaba Y, Yoshimoto T (2008) Prevalence and clinicoepidemiological features of moyamoya disease in Japan: findings from a nationwide epidemiological survey. Stroke 39:42–47
21. Kuroda S, Houkin K, Ishikawa T, Nakayama N, Iwasaki Y (2010) Novel bypass surgery for moyamoya disease using pericranial flap: its impacts on cerebral hemodynamics and long-term outcome. Neurosurgery 66:1093–1101, discussion 1101
22. Kuroda S, Houkin K, Kamiyama H, Abe H (2001) Effects of surgical revascularization on peripheral artery aneurysms in moyamoya disease: report of three cases. Neurosurgery 49:463–467, discussion 467–468
23. Miyamoto S (2004) Study design for a prospective randomized trial of extracranial-intracranial bypass surgery for adults with moyamoya disease and hemorrhagic onset—the Japan Adult Moyamoya Trial Group. Neurol Med Chir (Tokyo) 44:218–219
24. Miyamoto S, Akiyama Y, Nagata I, Karasawa J, Nozaki K, Hashimoto N, Kikuchi H (1998) Long-term outcome after STA-MCA anastomosis for moyamoya disease. Neurosurg Focus 5:e5
25. Morioka M, Hamada J, Todaka T, Yano S, Kai Y, Ushio Y (2003) High-risk age for rebleeding in patients with hemorrhagic moyamoya disease: long-term follow-up study. Neurosurgery 52:1049–1054, discussion 1054–1045
26. Mugikura S, Takahashi S, Higano S, Shirane R, Sakurai Y, Yamada S (2002) Predominant involvement of ipsilateral anterior and posterior circulations in moyamoya disease. Stroke 33:1497–1500
27. Mukawa M, Nariai T, Matsushima Y, Tanaka Y, Inaji M, Maehara T, Aoyagi M, Ohno K (2012) Long-term follow-up of surgically treated juvenile patients with moyamoya disease. J Neurosurg Pediatr 10: 451–456
28. Okada Y, Shima T, Nishida M, Yamane K, Yamada T, Yamanaka C (1998) Effectiveness of superficial temporal artery-middle cerebral artery anastomosis in adult moyamoya disease: cerebral hemodynamics and clinical course in ischemic and hemorrhagic varieties. Stroke 29:625–630
29. Phi JH, Wang KC, Cho BK, Lee MS, Lee JH, Yu KS, Hahm BJ, Kim SK (2011) Long-term social outcome in children with moyamoya disease who have reached adulthood. J Neurosurg Pediatr 8:303–309
30. Research Committee on the Pathology and Treatment of Spontaneous Occlusion of the Circle of Willis; Health Labour Sciences Research Grant for Research on Measures for Intractable Diseases (2012) Guidelines for diagnosis and treatment of moyamoya disease

(Spontaneous Occlusion of the Circle of Willis). Neurol Med Chir (Tokyo) 52:245–266
31. Scott RM, Smith JL, Robertson RL, Madsen JR, Soriano SG, Rockoff MA (2004) Long-term outcome in children with moyamoya syndrome after cranial revascularization by pial synangiosis. J Neurosurg 100:142–149
32. Starke RM, Komotar RJ, Hickman ZL, Paz YE, Pugliese AG, Otten ML, Garrett MC, Elkind MS, Marshall RS, Festa JR, Meyers PM, Connolly ES Jr (2009) Clinical features, surgical treatment, and long-term outcome in adult patients with moyamoya disease. Clinical article. J Neurosurg 111:936–942
33. Suzuki K, Sarti C, Tuomilehto J, Kutsuzowa T, Narva EV, Sivenius J, Salmi K, Kaarsalo E, Torppa J, Kuulasmaa K (1994) Stroke incidence and case fatality in Finland and in Akita, Japan: a comparative study. Neuroepidemiology 13:236–244
34. Takikawa S, Kamiyama H, Abe H, Mitsumori K, Tsuru M (1990) Hemodynamic evaluation of vascular reconstructive surgery for childhood moyamoya disease using single photon emission computed tomography. Neurol Med Chir (Tokyo) 30:389–395
35. Uchino H, Kuroda S, Hirata K, Shiga T, Houkin K, Tamaki N (2012) Predictors and clinical features of postoperative hyperperfusion after surgical revascularization for moyamoya disease: a serial single photon emission CT/positron emission tomography study. Stroke 43:2610–2616
36. Ulrich PT, Januschek E (2011) Revascularisation surgery and long-term follow-up in juvenile Moyamoya syndrome: a retrospective analysis. Acta Neurochir Suppl 112:39–43
37. Wakai K, Tamakoshi A, Ikezaki K, Fukui M, Kawamura T, Aoki R, Kojima M, Lin Y, Ohno Y (1997) Epidemiological features of moyamoya disease in Japan: findings from a nationwide survey. Clin Neurol Neurosurg 99(Suppl 2):S1–S5
38. Yoshida Y, Yoshimoto T, Shirane R, Sakurai Y (1999) Clinical course, surgical management, and long-term outcome of moyamoya patients with rebleeding after an episode of intracerebral hemorrhage: an extensive follow-up study. Stroke 30:2272–2276

Comment

The natural history of patients with moyamoya disease after revascularisation is still not well known. This manuscript deals with 56 children with a mean follow-up interval of 18.1 years; one ischemic stroke and three hemorrhages were observed. One of three hemorrhages was due to rupture of a small aneurysm at the posterior choroidal artery. These four events occurred after 13 years on average after revasculalisation surgery. The reviewer read this report with great interest as he had experience recently of an adult female patient (50 years old) who had suffered a huge hemorrhage in the right temporal lobe and right ventricle around 24 months after revascularisation surgery (bilateral STA-MCA bypass plus one STA-ACA bypass in one stage and patency of all the bypasses were confirmed with DSA). Whether revascularisation surgery prevents future hemorrhagic event or not—and, if yes, to what extent—has to be resolved. Interestingly, the hemorrhages reported in this paper and also in our patient were only in adults, occurring 18 years later and 1 year later respectively.

The mechanism of failure of prevention of hemorrhage that occurred so long after revascularisation surgery in the former case and so shortly after in the latter has also to be solved. Anyway, occurrence of hemorrhage seems to be associated with adulthood irrespective of revascularisation surgery.

By the way, fatal rerupture of the small intraventricular aneurysm of the posterior choroidal artery could have been prevented by its surgical removal, although some papers propose its shrinkage or disappearance by revascularisation.

Reference

Sadatoh A. Yonekawa Y, Morooka Y, Imakita T: A case of moyamoya disease with repeated intraventricular hemorrhage due to a ruptured pseudoaneurysm. No Shinkeigeka (1989)17:755-758 (Japanese, English abstract)

Yasuhiro Yonekawa

Zurich, Switzerland

Cerebrovascular Diseases

Received: January 8, 2013
Accepted: May 15, 2013
Published online: September 11, 2013

Cerebrovasc Dis 2013;36:155–157
DOI: 10.1159/000352065

Rapid Progression of Unilateral Moyamoya Disease in a Patient with a Family History and an *RNF213* Risk Variant

Yohei Mineharu[a, d] *Yasushi Takagi*[c] *Jun C. Takahashi*[c] *Hirokuni Hashikata*[e] *Wanyang Liu*[d] *Toshiaki Hitomi*[d] *Hatasu Kobayashi*[d] *Akio Koizumi*[d] *Susumu Miyamoto*[c]

[a]Division of Neuroendovascular Therapy, Institute of Biomedical Research and Innovation, and [b]Department of Neurosurgery, Kobe City Medical Center General Hospital Kobe, Departments of [c]Neurosurgery and [d]Health and Environmental Sciences, Kyoto University Graduate School of Medicine, Kyoto, and [e]Department of Neurosurgery, Kitano Hospital, Tazuke Kofukai Medical Research Institute, Osaka, Japan

Introduction

Moyamoya disease (MMD) is a progressive steno-occlusive vasculopathy that involves large intracranial arteries accompanied by moyamoya collaterals [1, 2]. It was demonstrated that the p.R4810K missense variant (rs 112735431) in the *RNF213* gene on the 17q25.3 locus [3–5] increases susceptibility to MMD in East Asian populations [6]. Genetic diagnosis enabled us to find presymptomatic patients with MMD.

Case Report

A 36-year-old woman, who had no past medical history, received MRI screening examination to check for MMD because her mother and her aunt had the disease. The initial examination in August 2005 showed no apparent intracranial arterial stenosis (fig. 1a). One year later, she received the second MRI scan, which showed proximal right middle cerebral artery (MCA) occlusion (fig. 1b). Conventional angiography confirmed MCA occlusion with minor moyamoya collaterals at the base of the brain (fig. 1c). No stenosis was observed on the contralateral side. Despite rapid progression of the arterial occlusion, the patient did not develop any neurological symptoms or ischemic brain lesions on MRI. She was conservatively followed up by annual MRI examinations without surgical intervention. The occlusive lesion has remained stable for 6 years without any progression.

The patient and her family members, including unaffected members, received both genetic testing for *RNF213* and MRI examination in 2005; the family is pedigree 18 in our previous paper [6]. Sequencing of *RNF213* in the patient's mother and aunt revealed two haplotypes carrying p.R4810K: allele A_2, which is common among patients with MMD, and allele A_1, which is rare among patients with MMD [6]. The patient inherited an A_1 allele for p.R4810K (fig. 1d). On the other hand, her elder and younger sisters inherited an A_2 allele from their mother for p.R4810K, and no arterial stenosis was identified in either the initial or annual follow-up MRI examinations.

Ethical approval for this study was given by the Institutional Review Board and Ethics Committee of the Kyoto University School of Medicine, Kyoto University, Japan.

Discussion

Due to incomplete penetrance of the p.R4810K variant, *RNF213* is considered to be a susceptibility gene and other genetic or environmental factors may be associated with MMD. However, the genome-wide linkage and association analysis only showed a significant signal in *RNF213* on 17q25.3, indicating that other genetic factors have a much lower effect as compared with *RNF213* [3, 6]. p.R4810K or other mutations in *RNF213* were observed in all familial cases of MMD including Japanese, Korean and European populations and p.R4810K was associated with an increased risk of MMD with an odds ratio of as much as 338.9 for a Japanese population [6], which was confirmed in independent studies [5, 7]. These results indicate that p.R4810K screening would be a most appropriate approach to identify asymptomatic patients, especially those who have a family history of MMD.

In the present study, a 36-year-old woman, who was positive for *RNF213*, had de novo progression of unilateral MMD within only a year. In the past reports, a 59-year-old woman showed de novo progression of bilateral MMD within a 5-year interval and a 46-year-old woman developed unilateral MMD between 2004 and 2009 [8, 9]. Albeit not adult cases, Amlie-Lefond et al. [10] reported a 3-month-old patient with MMD and reviewed other 8 cases of early infancy before the age of 1 year, suggesting that MMD can develop very rapidly. Therefore, frequent follow-ups by MRI should be recommended for those who were diagnosed as having genetic risk factors for MMD.

Although the elder and younger sisters of the patient had the p.R4810K variant, they have not developed MMD. Since they may develop MMD several years later, a close follow-up is necessary. Alternatively, the discordant phenotype of the sisters may represent allelic differences between A_1 and A_2 [6]. The patient and the affected mother and aunt share p.R4810K on the same allele (A_1), whereas the unaffected sisters have A_2, suggesting a possibility that the 5′ portion of *RNF213* may have a modifier effect on the steno-occlusive phenotype. Still another possibility includes environmental factors, which may affect the pene-

1015–9770/13/0362–0155$38.00/0

E-Mail karger@karger.com
www.karger.com/ced

Yohei Mineharu
Division of Neuroendovascular Therapy
Institute of Biomedical Research and Innovation
2-2, Minatojima-Minamimachi, Chuo-ku, Kobe 650-0047 (Japan)
E-Mail mineharu@kuhp.kyoto-u.ac.jp

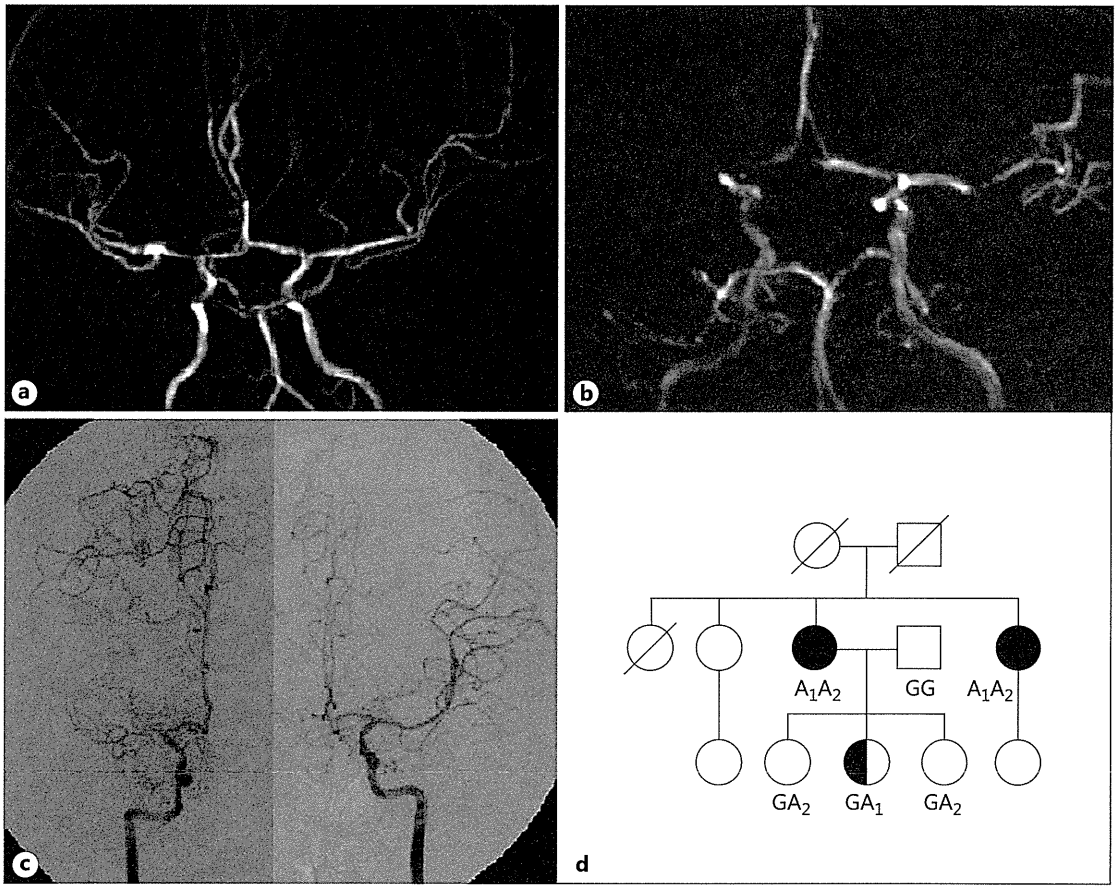

Fig. 1. a Initial magnetic resonance angiography (MRA; 1.5 tesla) of the patient. No arterial stenosis was observed. **b** Second MRA (1.5 tesla) study 1 year after the initial examination showed disruption of the right MCA. **c** Digital subtraction angiography following the second MRA examination revealed right MCA occlusion and the formation of collateral circulation at the base of the brain. Distal MCA was filled with contrast media via collaterals. **d** The family with familial MMD. Filled symbols indicate patients with MMD; half-filled symbols, patients with unilateral MMD; circles, women; squares, men; crossed symbols, deceased people. The half-filled symbol in the third generation represents the 36-year-old patient in this report. Genotypes of the risk allele of *RNF213* are shown. GG represents wild type; GA, homozygote for the risk variant, AA heterozygote for the risk variant. Numbers are attached to the variant 'A' to discriminate the two different alleles. The patient and her affected mother and aunt share the risk variant on the same allele (A_1).

trance, although we could not identify any environmental differences between the affected patient and the unaffected sisters. The patient had one A allele (heterozygote) and developed unilateral MMD, whereas her mother and aunt had two A alleles (homozygote) and developed bilateral MMD. Miyatake et al. [7] reported that the number of risk alleles in *RNF213* is associated with earlier age at onset and a severe form of the disease. Bilateral progression may also be associated with the number of risk alleles. Further follow-ups and investigations are warranted for this family.

Acknowledgements

This work was partly supported by grants from the Ministry of Education, Science, Sports and Culture of Japan (Kiban Kenkyu A: 14207016 and 22249020, S: 17109007, Tokutei Kenkyu: 15012231, 16012232, 17019034 and 18018022) and Research Committee on Moyamoya Disease of the Ministry of Health, Labor and Welfare, Japan.

Disclosure Statement

None.

References

1 Guidelines for diagnosis and treatment of moyamoya disease (Spontaneous Occlusion of the Circle of Willis). Neurol Med Chir (Tokyo) 2012; 52:245–266.
2 Czabanka M, Peña-Tapia P, Schubert GA, Heppner FL, Martus P, Horn P, et al: Proposal for a new grading of Moyamoya disease in adult patients. Cerebrovasc Dis 2011;32:41–50.
3 Mineharu Y, Liu W, Inoue K, Matsuura N, Inoue S, Takenaka K, et al: Autosomal dominant moyamoya disease maps to chromosome 17q25.3. Neurology 2008;70:2357–2363.
4 Mineharu Y, Takenaka K, Yamakawa H, Inoue K, Ikeda H, Kikuta K-I, et al: Inheritance pattern of familial moyamoya disease: autosomal dominant mode and genomic imprinting. J Neurol 2006;77:1025–1029.
5 Kamada F, Aoki Y, Narisawa A, Abe Y, Komatsuzaki S, Kikuchi A, et al: A genome-wide association study identifies RNF213 as the first Moyamoya disease gene. J Hum Genet 2010;56:1–7.
6 Liu W, Morito D, Takashima S, Mineharu Y, Kobayashi H, Hitomi T, et al: Identification of RNF213 as a susceptibility gene for moyamoya disease and its possible role in vascular development. PLoS One 2011; 6:e22542.
7 Miyatake S, Miyake N, Touho H: Homozygous c.14576G>A variant of RNF213 predicts early-onset and severe form of moyamoya disease. Neurology 2012;78:803–810.
8 Fukaya R, Yoshida K, Akiyama T, Kawase T: De novo development of moyamoya disease in an adult female. Case report. J Neurosurg 2009;111: 943–946.
9 Shimoda Y, Fujimura M, Inoue T, Shimizu H, Tominaga T: Temporal profile of de novo development of moyamoya vasculopathy in an adult. Neurol Med Chir (Tokyo) 2012;52:339–342.
10 Amlie-Lefond C, Bernard TJ, Sébire G, Friedman NR, Heyer GL, Lerner NB, et al: Predictors of cerebral arteriopathy in children with arterial ischemic stroke: results of the International Pediatric Stroke Study. Circulation 2009;119:1417–1423.

PLOS | ONE

RESEARCH ARTICLE

Voxel Based Analysis of Surgical Revascularization for Moyamoya Disease: Pre- and Postoperative SPECT Studies

Yasutaka Fushimi[1]*, Tomohisa Okada[1], Yasushi Takagi[2], Takeshi Funaki[2], Jun C. Takahashi[2], Susumu Miyamoto[2], Kaori Togashi[1]

1 Department of Diagnostic Imaging and Nuclear Medicine, Kyoto University Graduate School of Medicine, Kyoto, 606–8507, Japan, **2** Department of Neurosurgery, Kyoto University Graduate School of Medicine, Kyoto, 606–8507, Japan

* yfushimi@kuhp.kyoto-u.ac.jp

OPEN ACCESS

Citation: Fushimi Y, Okada T, Takagi Y, Funaki T, Takahashi JC, Miyamoto S, et al. (2016) Voxel Based Analysis of Surgical Revascularization for Moyamoya Disease: Pre- and Postoperative SPECT Studies. PLoS ONE 11(2): e0148925. doi:10.1371/journal.pone.0148925

Editor: Gustavo Pradilla, Emory University School of Medicine, UNITED STATES

Received: July 8, 2015

Accepted: December 16, 2015

Published: February 11, 2016

Data Availability Statement: There is a procedure at the authors' institution through which interested researchers can apply to access underlying data. Data access requests are reviewed by the Kyoto University Graduate School and Faculty of Medicine Ethics Committee. Please contact the Corresponding Author for details related to initiating this process.

Funding: This study was supported by JSPS KAKENHI grant number 25461815, https://www.jsps.go.jp/english/index.html, and Nihon Medi-Physics, http://www.nmp.co.jp/eng/nuclear_medicine/cooperation.html. The funders had no role in study

Abstract

Moyamoya disease (MMD) is a chronic, progressive, cerebrovascular occlusive disease that causes abnormal enlargement of collateral pathways (moyamoya vessels) in the region of the basal ganglia and thalamus. Cerebral revascularization procedures remain the preferred treatment for patients with MMD, improving the compromised cerebral blood flow (CBF). However, voxel based analysis (VBA) of revascularization surgery for MMD based on data from pre- and postoperative data has not been established. The latest algorithm called as Diffeomorphic Anatomical Registration Through Exponentiated Lie Algebra (DARTEL) has been introduced for VBA as the function of statistical parametric mapping (SPM8), and improved registration has been achieved by SPM8 with DARTEL. In this study, VBA was conducted to evaluate pre- and postoperative single photon emission computed tomography (SPECT) images for MMD by SPM8 with DARTEL algorithm, and the results were compared with those from SPM8 without DARTEL (a conventional method). Thirty-two patients with MMD who underwent superficial temporal artery-middle cerebral artery (STA-MCA) bypass surgery as the first surgery were included and all patients underwent pre- and postoperative 3D T1-weighted imaging and SPECT. Pre- and postoperative SPECT images were registered to 3D T1-weighted images, then VBA was conducted. Postoperative SPECT showed more statistically increased CBF areas in the bypassed side cerebral hemisphere by using SPM8 with DARTEL (58,989 voxels; $P<0.001$), and increased ratio of CBF after operation was less than 15%. Meanwhile, postoperative SPECT showed less CBF increased areas by SPM8 without DARTEL. In conclusion, VBA was conducted for patients with MMD, and SPM8 with DARTEL revealed that postoperative SPECT showed statistically significant CBF increases over a relatively large area and with at most 15% increase ratio.

design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing Interests: This study was funded by JSPS KAKENHI grant number 25461815 and Nihon Medi-Physics. There are no patents, products in development or marketed products to declare. This does not alter the authors' adherence to all the PLOS ONE policies on sharing data and materials as detailed online in the guide for authors.

Introduction

Moyamoya disease (MMD) is a chronic, progressive, cerebrovascular occlusive disease that causes abnormal enlargement of collateral pathways (moyamoya vessels) in the region of the basal ganglia and thalamus. Cerebral revascularization procedures remain the preferred treatment for patients with MMD [1], improving the compromised cerebral blood flow (CBF), reducing ischemic attacks, and resulting in sufficiently good long-term results [2]. However, in spite of established revascularization procedures, the method for evaluating the efficacy of revascularization surgery for MMD based on data from pre- and postoperative data has not been established.

Postoperative cerebral hyperperfusion syndrome has been considered to be less common in patients with MMD, because relatively low-flow revascularization can usually be attained surgically for MMD [3–5]. Revascularization surgery in patients with MMD carries a low risk, is effective at preventing future ischemic events, and improves quality of life [6, 7]. However, a recent study reported that cerebral hyperperfusion syndrome after superficial temporal artery (STA)-middle cerebral artery (MCA) bypass occurs more frequently in patients with MMD than in those with arteriosclerotic disease, with a diagnostic criteria of qualitative observation of focal intense CBF increase [8]. It is evident that favorable postoperative CBF increase as the effect of STA-MCA bypass should be differentiated from unwanted hyperperfusion syndrome.

Given these considerations, objective measurements are required to confirm the potential advantages of revascularization surgery, and many quantitative approach has been conducted [9]. On the contrary, voxel based analysis (VBA) is now widely performed, especially for magnetic resonance imaging (MRI) analysis. This technique enables fully automatic processing of images and is considered highly objective. Postoperative evaluation of MMD has been reported with dynamic susceptibility-weighted perfusion MRI [10], arterial spin labeling [11], computed tomography perfusion [12], and single-photon emission computed tomography (SPECT) [13], but those studies used region-of-interest analysis, and no VBA study has previously been conducted probably due to misregistration associated with postoperative brain shift. VBA for CBF changes for ischemia in the internal carotid artery area after bypass surgery was reported previously with relatively old algorithm [14]. With the advent of new algorithm such as the Diffeomorphic Anatomical Registration Through Exponentiated Lie Algebra (DARTEL), which has been introduced for VBA as the function of statistical parametric mapping (SPM8), improved registration accuracy has been brought to realization [15], VBA with the latest algorithm should be verified in pre- and postoperative images. This study was conducted to evaluate pre- and postoperative single photon emission computed tomography (SPECT) images for MMD by SPM8 with DARTEL algorithm, and the results were compared with those from SPM8 without DARTEL (a conventional method).

In this study, SPECT images of pre- and post-revascularization surgery for MMD were analyzed by VBA with DARTEL algorithm, and the results were compared with those from VBA without DARTEL (conventional method).

Materials and Methods

Patients

This retrospective study was approved by the institutional review board "Kyoto University Graduate School and Faculty of Medicine, Ethics Committee" and the need for written informed consent was waived. Patient information was anonymized and de-identified prior to analysis. Participants comprised 33 consecutive adult patients with MMD who underwent first STA-MCA bypass surgery at our institute between January 2010 and June 2013. The diagnosis

of MMD was made in accordance with published guidelines for bilateral MMD [16, 17]. Patients with typical occlusive findings in the terminal portion of a unilateral internal carotid artery, which was diagnosed as probable MMD, were also included in this study (unilateral MMD), because asymmetry in the progression of stenosis is relatively common among patients with unilateral MMD [18]. Patients with autoimmune disease, Down syndrome or neurofibromatosis were excluded. Patients younger than 18 years old were also excluded, because age-related changes in cerebral blood flow (CBF) might occur, and STA-MCA bypass with encephalo-myosynangiosis was conducted for child patients instead of STA-MCA bypass surgery only [19]. Patients who had undergone previous bypass surgery or other operation were also excluded. Patients with intracranial hemorrhage and patients with cerebral infarction of greater than 1/3 of MCA territory, or major territorial infarction of anterior or posterior cerebral infarction were also excluded.

All patients in this study underwent preoperative imaging with both SPECT and MRI. All patients underwent STA-MCA bypass as the first surgery and the branches of the STA were anastomosed to the central branch of MCA by end-to-side anastomosis [20]. Postoperative SPECT and MRI were obtained on around postoperative day 5. All but 1 patient with postoperative focal cerebral infarction were included for comparisons between pre- and postoperative SPECT, and this patient was excluded from the study.

Imaging Study

SPECT was acquired at resting state using a 2-head rotating gamma camera (Infinia; GE Medical Systems, Milwaukee, USA) with an extended low-energy general-purpose collimator. Patients were asked to keep the eyes closed during scanning. A bolus of N-isopropyl-p-[(123) I]-iodoamphetamine (^{123}I-IMP) (167 MBq) with 10 ml of normal saline was administered intravenously at the beginning of image acquisition. Data were acquired in a 64 × 64 matrix through a 120° rotation at angle intervals of 4°. Total imaging time was 30 min. Spatial resolution at the center of view was 9.9 mm in full-width at half-maximum (FWHM) activity. Transverse reconstruction with Ramp and Butterworth filters (cutoff of 0.5 cycles/pixel and order 10), and attenuation correction using the method of Chang (0.07/cm) were applied. FWHM of the collimator was 10 mm.

Pre- and postoperative MR scans included 3-dimensional (3D) T1-weighted imaging, diffusion weighted imaging, 2D Fluid Attenuated Inversion Recovery (FLAIR) imaging and MR angiography using 3-T MR units (Magnetom Trio and Magnetom Skyra; Siemens, Erlangen, Germany), using 32-channel head coil. The parameter of 3D T1-weighted imaging was as follows: magnetization prepared rapid acquisition with gradient echo (MPRAGE) repetition time 1900 msec, echo time 2.58 msec, inversion time 900 msec, flip angle 9 degrees, field of view 230 × 230 mm, acquisition matrix 256 × 256, pixel size 0.90 × 0.90 mm, slice thickness 0.9 mm.

Postimaging Analysis

Because the patients underwent bypass surgery on either the left or right side, the operated side was made to appear as the right side by horizontally flipping left-side patient images. Thus all SPECT data could be dealt with as data from patients with right STA-MCA bypass surgery. The count of each voxel was normalized by the total count for the brain (proportional scaling of the global mean to 100). Pre- and postoperative IMP-SPECT images were co-registered to corresponding 3D T1-weighted images using statistical parametric mapping (SPM8) software (Wellcome Department of Imaging Neuroscience, University College London, UK) implemented on MATLAB 2013b (Mathworks, Natick, Massachusetts, USA). The 3D T1 images were then segmented into gray matter (GM), white matter (WM) and cerebrospinal fluid

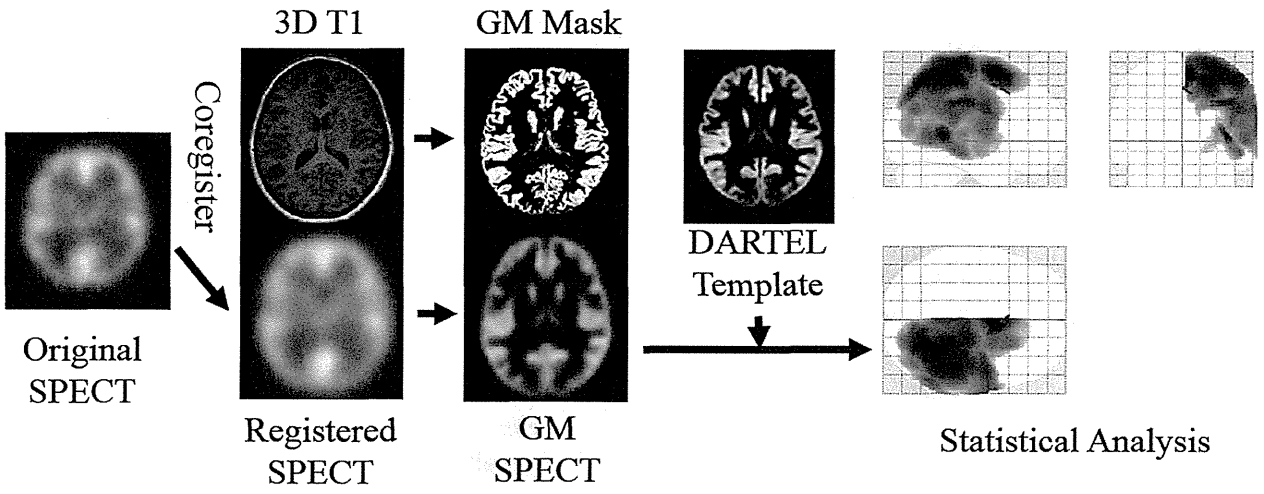

Fig 1. The flowchart of postimaging process of VBA. In the first place, horizontally flipping was conducted for the SPECT data with left side operation in order to deal with all the SPECT data as that with right STA-MCA bypass surgery. After global mean normalization was performed, pre- and postoperative IMP-SPECT images were co-registered to corresponding 3D T1-weighted images. The 3D T1 images were then segmented into gray matter (GM), white matter (WM) and cerebrospinal fluid (CSF) space using new segmentation tool and DARTEL import files were created for each patient. GM segment of coregistered SPECT (GM-SPECT) was created. Then, DARTEL template was generated with DARTEL import files. In the last place, non-linear warping of all GM images including GM-SPECT to MNI space was conducted with 2 methods as follows, with DARTEL by using flow field, or without DARTEL (a conventional method, which has been used in the previous literatures).

doi:10.1371/journal.pone.0148925.g001

(CSF) space using new segmentation tool in SPM8, and DARTEL import files were created for each patient during this process [21]. A DARTEL template was generated from the entire image dataset using DARTEL import files. In the next step, the GM segment of coregistered SPECT (GM-SPECT) was created. Non-linear warping of all GM images to the Montreal Neurological Institute (MNI) space (http://www.mni.mcgill.ca/) was conducted with 2 methods as follows, (a) non-linear warping was conducted with DARTEL template and flow fields which store the deformation information, (b) non-linear warping was conducted without DARTEL (a conventional method, which has been used in the previous literatures), then the normalization parameters derived were applied to GM-SPECT, respectively. Spatially normalized GM-SPECT images were subsequently smoothed using an isotopic Gaussian kernel with a 12-mm full-width at half-maximum (Fig 1).

Postimaging Analysis

ROI analysis was performed for each dataset of pre- and postoperative SPECT (GM-SPECT) in order to validate DARTEL method [22]. ROIs of bilateral precentral, central and parietal areas were created (Fig 2). Asymmetry index was created for the bypass side by using bilateral ROIs, then pre- and post-operative ROIs were compared.

Statistical Analysis

Paired *t*-test was performed between pre- and postoperative GM-SPECT images voxel-by-voxel, and postoperative CBF increase were investigated. In SPM analysis, the P value threshold was 0.001 at the voxel level, and clusters were considered as significant when falling below a cluster-corrected p (FDR) = 0.05. Increased ratios (%) and decrease ratios (%) of postoperative CBF compared with preoperative CBF at areas of significant CBF increase were calculated and shown by using the xjView toolbox (http://www.alivelearn.net/xjview).

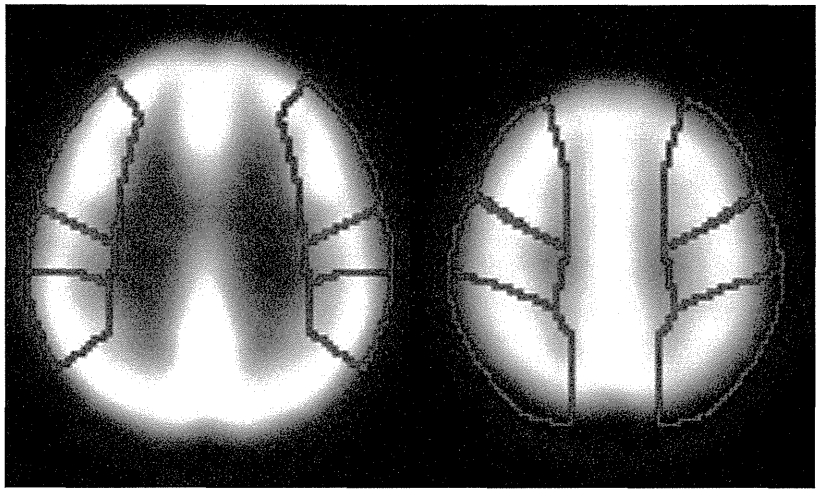

Fig 2. ROIs of bilateral precentral, central and parietal area. ROIs of bilateral precentral, central and parietal areas were created, and the same ROIs were applied for all cases.

doi:10.1371/journal.pone.0148925.g002

Paired *t*-test was also performed ROI values between pre- and postoperative GM-SPECT images. P value threshold < 0.05 was considered as significant.

Results

In total, 32 hemispheres of 32 patients (9 males, 23 females; mean age, 38.2 years; range, 18–52 years) underwent STA-MCA anastomosis (bilateral MMD, n = 24; unilateral MMD, n = 8) (Table 1). The contralateral hemisphere was operated later in 12 of the 24 patients with bilateral MMD, but only the hemisphere of first surgery was analyzed in this study. Postoperative SPECT was conducted at a mean of 5.1 days (range, 2–11 days) after surgery.

SPM8 with DARTEL

SPM results showed that significant postoperative CBF increases in the right cerebral hemisphere or bypassed side (58,989 voxels; Fig 3A), as the side of operation was depicted as the right side by flipping the images, as necessary. These findings suggest that increased CBF due to STA-MCA anastomosis at the central branch of MCA appears not only at the site of anastomosis, but also in the wider area.

Increased ratio (%) at the area of significantly increased CBF on postoperative SPECT compared with preoperative SPECT is shown in Fig 4A, and increased ratio was less than 15%.

Decreased ratio (%) at the area of significantly decreased CBF on postoperative SPECT compared with preoperative SPECT was shown in contralateral hemisphere to the STA-MCA bypass site (Fig 5A). Since global mean normalization to the value of 100 was conducted for all SPECT data, ipsilateral CBF increase in wide area is likely to cause pseudo CBF decrease in the contralateral side. Visual inspection was performed for all the individual SPECT data to make assurance, and no misregistration was present in the contralateral hemisphere.

Table 1. Profile of Patients Included in the Comparison of Pre- and Postoperative SPECT.

Initial Bypass Surgery	Bilateral MMD	Unilateral MMD
Right	16	6
Left	8	2

doi:10.1371/journal.pone.0148925.t001

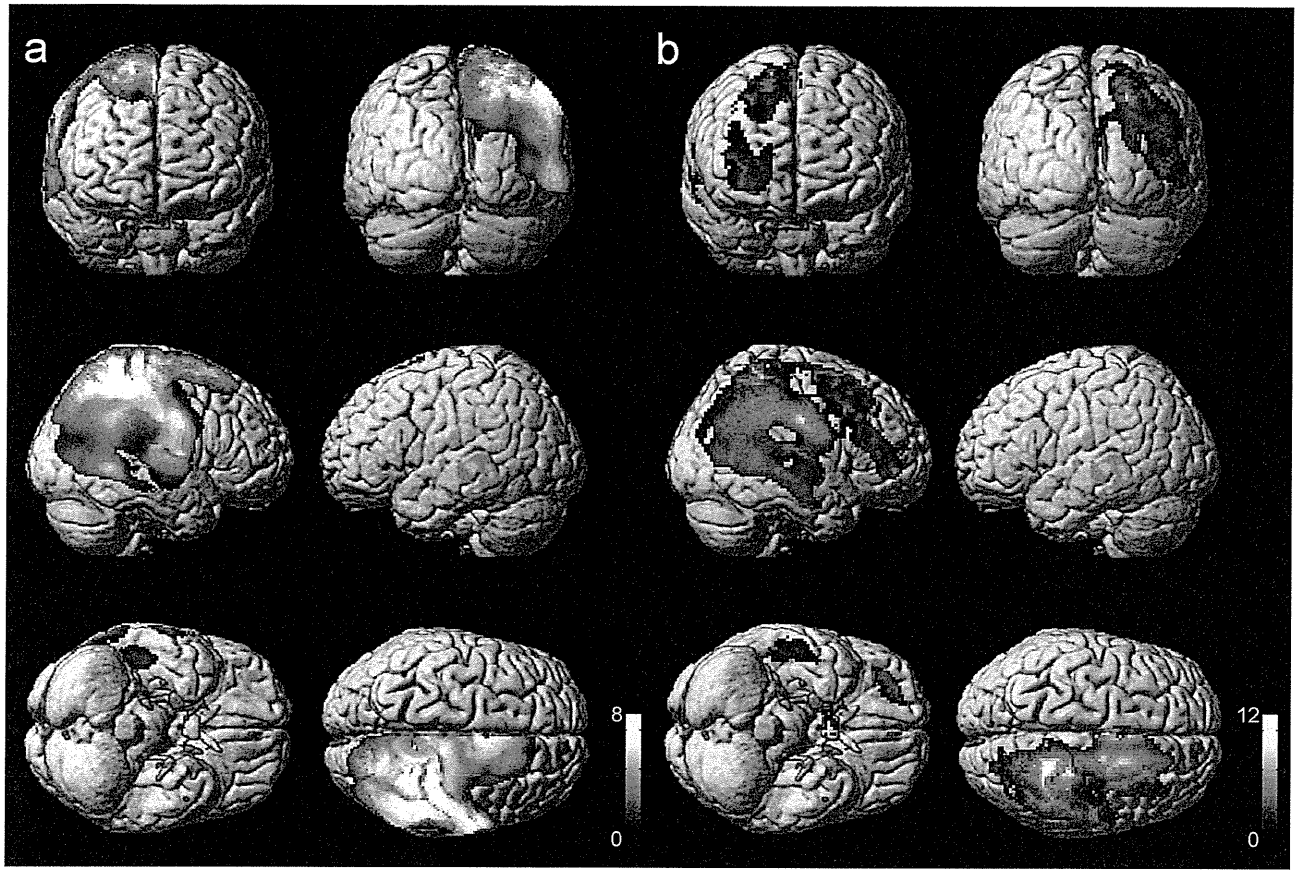

Fig 3. Postoperative CBF increased areas displayed in surface rendered images. Surface rendered images showed that significant postoperative CBF increased areas (58,989 voxels) in the right cerebral hemisphere or bypassed side by using SPM8 with DARTEL (a). These findings suggest that increased CBF due to STA-MCA anastomosis at the central branch of MCA appears not only at the site of anastomosis, but also in the wider area. Meanwhile, less significant areas (21,134 voxels) were shown by SPM8 without DARTEL (a conventional method) (b).

doi:10.1371/journal.pone.0148925.g003

SPM8 without DARTEL (A Conventional Method)

SPM results showed that significant postoperative CBF increases in the right cerebral hemisphere or bypassed side (21,134 voxels; Fig 3B).

Increased ratio (%) at the area of significantly increased CBF on postoperative SPECT compared with preoperative SPECT is shown in Fig 4B. CBF increased areas were located in more medial regions, and relatively smaller than those with DARTEL. Increased ratio was less than 15%.

Decreased ratio (%) at the area of significantly decreased CBF on postoperative SPECT compared with preoperative SPECT was shown on the surface of the ipsilateral hemisphere of bypass site (Fig 5B). Apparent misregistration was not detected by visual inspection, however, slight medial shift of postoperative cerebral cortices derived from operation procedure compared with the preoperative MRI might induce mismatched comparison in MNI space between pre- and postoperative SPECT due to less accurate normalization to MNI space compared with DARTEL, because most counts of SPECT are derived from cerebral cortices. No decreased CBF areas were detected in the contralateral hemisphere, which suggests that decreased CBF areas in the operated hemisphere compensate CBF increase from the viewpoint of global mean normalization method.

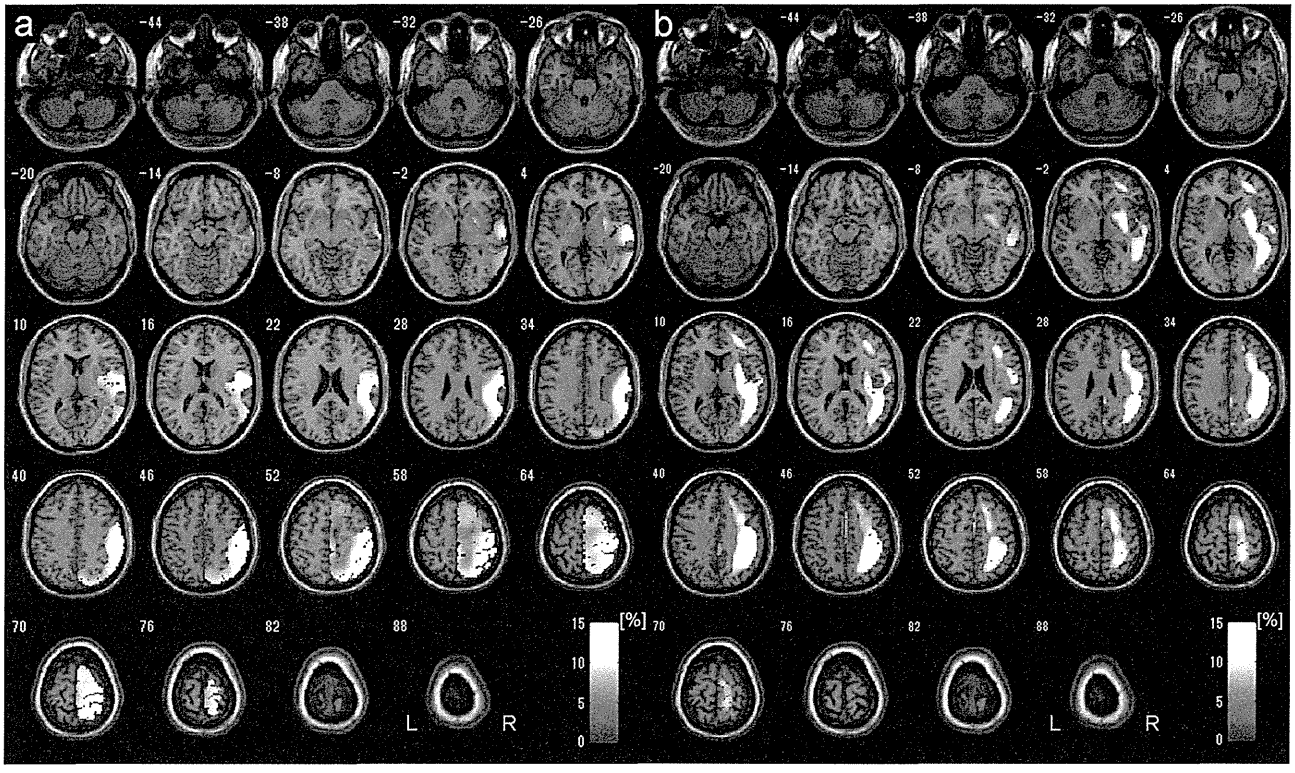

Fig 4. Postoperative CBF increased areas displayed in transverse sections. Increased ratio (%) at the area of significantly increased CBF on postoperative SPECT compared with preoperative SPECT is shown by using SPM8 with DARTEL (a) and SPM8 without DARTEL (a conventional method) (b). Images were reconstructed shown in 6mm thickness and the increased ratio was less than 15% in both methods. SPM8 with DARTEL revealed larger CBF increased areas (a), on the contrary, the conventional method showed relatively smaller CBF increased areas which were located in more medial regions compared with those with DARTEL.

doi:10.1371/journal.pone.0148925.g004

The summary of statistically important CBF changes after bypass surgery is shown in Table 2.

ROI Analysis

ROI analysis also showed post-operative increase as shown as below and Fig 6. Results of ROI analysis were as follows: precentral 3.61 ± 1.23 ($p = 0.007$), central 6.57 ± 1.28 ($p<0.001$), and parietal 9.34 ± 1.55 (%) ($p<0.001$) (Fig 6).

Discussion

Both VBA by using SPM8 with DARTEL and SPM8 without DARTEL showed ipsilateral CBF increase in the STA-MCA bypass side, and SPM8 with DARTEL showed larger CBF increased area (Figs 3 and 4). On the contrast, SPM8 without DARTEL showed significant ipsilateral CBF decrease at the surface of anastomosis site, which may suggest the focal misregistration probably due to postoperative changes in the images (Fig 5B). VBA for comparison between preoperative and postoperative SPECT successfully demonstrated CBF increase after STA-MCA bypass surgery, and DARTEL technique is considered to be robust against postoperative brain shift changes associated with STA-MCA bypass surgery.

Our study demonstrated a significant postoperative increase in a relatively large area of ipsilateral MCA territory in the operated hemisphere compared with preoperative SPECT, which

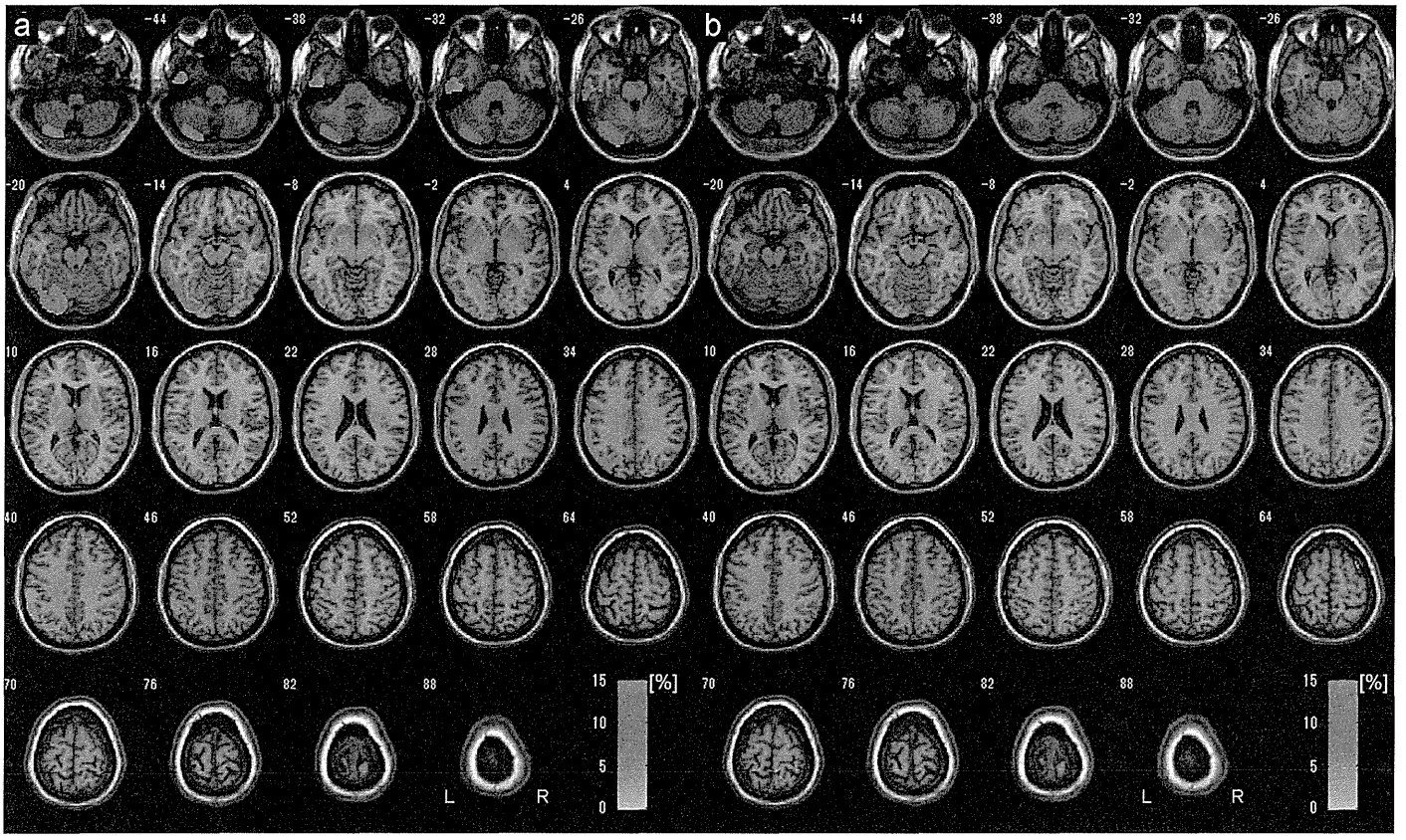

Fig 5. Postoperative CBF decreased areas displayed in transverse sections. Decreased ratio (%) at the area of significantly decreased CBF on postoperative SPECT compared with preoperative SPECT was shown in contralateral hemisphere to the STA-MCA bypass site by SPM8 with DARTEL (a). Since global mean normalization to the value of 100 was conducted for all SPECT data, ipsilateral CBF increase in wide area is likely to cause pseudo CBF decrease in the contralateral site. The conventional analysis showed significant decreased CBF areas on the surface of the ipsilateral hemisphere of bypass site (b). No decreased CBF areas were detected in the contralateral hemisphere, which suggests that decreased CBF areas in the operated hemisphere compensate CBF increase from the viewpoint of global mean normalization method.

doi:10.1371/journal.pone.0148925.g005

demonstrated the therapeutic effect of direct STA-MCA bypass. Hyperperfusion syndrome was originally defined as a $\geq$100% increase from baseline without any time limit, to avoid missing late-onset hyperperfusion syndrome [23]. Qualitative IMP-SPECT has previously been used to demonstrate that radiological hyperperfusion occurs in 50% of patients with MMD after surgery using inner control of MCA/ipsilateral cerebellum ratio [13]. Our results with DARTEL technique suggest that moderate CBF increase at most 15% compared with

Table 2. Summarys of Statistically Important CBF Changes after Bypass Surgery.

	CBF Increase		CBF Decrease	
	SPM8 with DARTEL	SPM8 without DARTEL (A Conventional Method)	SPM8 with DARTEL	SPM8 without DARTEL (A Conventional Method)
Operated Side	58989	21134	0	19230
Contralateral Side	0	0	14502	0

Note that the unit is voxels.

doi:10.1371/journal.pone.0148925.t002

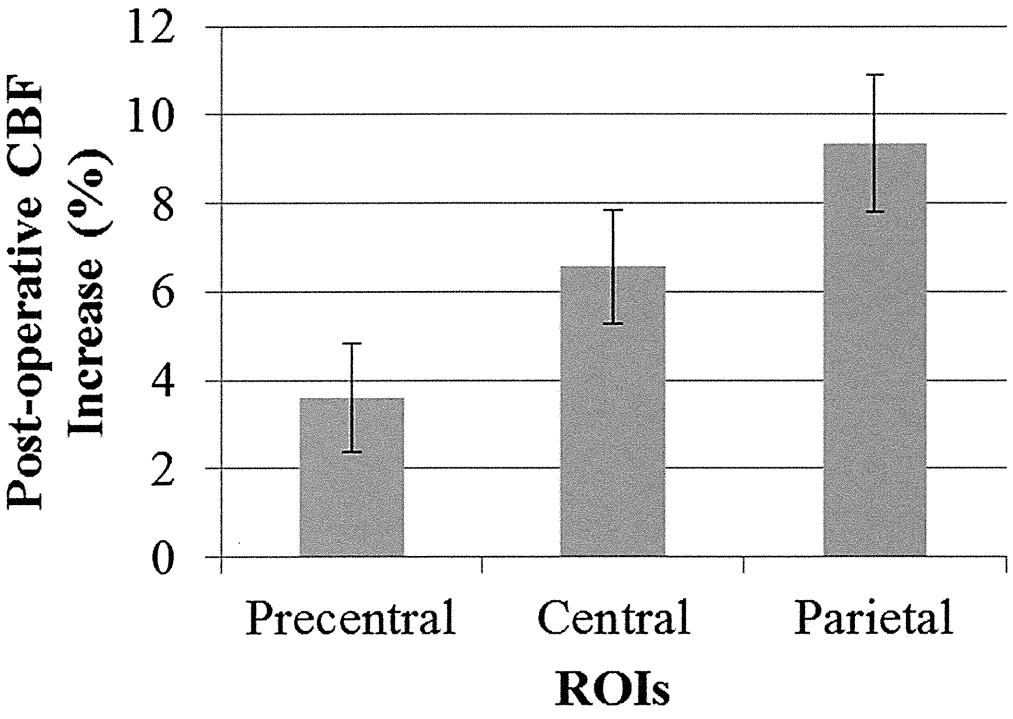

Fig 6. ROI analysis for post-operative CBF increase. Post operative CBF increase (%) at each ROI of bypass side are shown. The area near to the STA-MCA anastomosis showed higher increase ratio. Error bar represents standard errors.

doi:10.1371/journal.pone.0148925.g006

preoperative SPECT occur due to improved blood supply from STA-MCA bypass in the post-operative state (Fig 4A).

Bypass surgery has been considered to play two important roles in MMD. One is to salvage misery perfusion, and the other is to reduce the fragile collateral vessels known as moyamoya vessels, because rupture of moyamoya vessels is thought to cause cerebral hemorrhage in MMD [6, 24]. The actual reductions in the risk of cerebral hemorrhage by STA-MCA bypass have been unclear [25], however, a recent study revealed that extracranial–intracranial bypass improved patient prognosis compared with non-surgical treatment, suggesting the preventive effect of direct bypass against rebleeding [26].

In this study, analysis was conducted using VBA with DARTEL technique, which provides improved registration accuracy compared with conventional VBA methods which had been used in the previous literatures [27]. Bypass surgery caused postoperative changes, but using anatomical MRI of patients with MMD and creating a template specific to patients with MMD by using DARTEL process based on the flow filed storing deformation information may allow precise registration and normalization for statistical comparison [15].

Several limitations to this study must be considered. First, the influence of bypass site on CBF has not been clarified. Slight brain shift might lead to comparisons between WM on pre-operative SPECT and GM on postoperative SPECT; as a result, only GM on SPECT was analyzed in this study. Second, a correlation analysis with clinical data and individual patient analysis were not performed in this study. Since VBA has been less common in perioperative images, and more verification study might be necessary to establish VBA of longitudinal analysis for pre- and postoperative STA-MCA analysis. Third, cerebrovascular reserve was not analyzed in this study. Fourth, long-term follow-up exams were not included in this study.

In conclusion, VBA was conducted for patients with MMD, and SPM8 with DARTEL revealed that postoperative SPECT showed statistically significant CBF increases over a relatively large area and with at most 15% increase ratio.

Author Contributions

Conceived and designed the experiments: YF TO. Performed the experiments: YT TF JCT SM. Analyzed the data: YF TO. Contributed reagents/materials/analysis tools: YF. Wrote the paper: YF TO KT.

References

1. Pandey P, Steinberg GK. Neurosurgical advances in the treatment of moyamoya disease. Stroke; a journal of cerebral circulation. 2011; 42(11):3304–10. doi: 10.1161/STROKEAHA.110.598565 PMID: 21980214.
2. Miyamoto S, Akiyama Y, Nagata I, Karasawa J, Nozaki K, Hashimoto N, et al. Long-term outcome after STA-MCA anastomosis for moyamoya disease. Neurosurgical focus. 1998; 5(5):e5. PMID: 17112208.
3. Fujimura M, Kaneta T, Mugikura S, Shimizu H, Tominaga T. Temporary neurologic deterioration due to cerebral hyperperfusion after superficial temporal artery-middle cerebral artery anastomosis in patients with adult-onset moyamoya disease. Surg Neurol. 2007; 67(3):273–82. Epub 2007/02/27. doi: S0090-3019(06)00769-5 [pii] doi: 10.1016/j.surneu.2006.07.017 PMID: 17320638.
4. 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:S142–5. Epub 1997/12/31. PMID: 9409425.
5. Sakamoto T, Kawaguchi M, Kurehara K, Kitaguchi K, Furuya H, Karasawa J. Risk factors for neurologic deterioration after revascularization surgery in patients with moyamoya disease. Anesth Analg. 1997; 85(5):1060–5. Epub 1997/11/14. PMID: 9356100.
6. Guzman R, Lee M, Achrol A, Bell-Stephens T, Kelly M, Do HM, et al. Clinical outcome after 450 revascularization procedures for moyamoya disease. Clinical article. J Neurosurg. 2009; 111(5):927–35. doi: 10.3171/2009.4.JNS081649 PMID: 19463046.
7. Han JS, Abou-Hamden A, Mandell DM, Poublanc J, Crawley AP, Fisher JA, et al. Impact of extracranial-intracranial bypass on cerebrovascular reactivity and clinical outcome in patients with symptomatic moyamoya vasculopathy. Stroke; a journal of cerebral circulation. 2011; 42(11):3047–54. doi: 10.1161/STROKEAHA.111.615955 PMID: 21903951.
8. Fujimura M, Shimizu H, Inoue T, Mugikura S, Saito A, Tominaga T. Significance of focal cerebral hyperperfusion as a cause of transient neurologic deterioration after extracranial-intracranial bypass for moyamoya disease: comparative study with non-moyamoya patients using N-isopropyl-p-[(123)I] iodoamphetamine single-photon emission computed tomography. Neurosurgery. 2011; 68(4):957–64; discussion 64–5. doi: 10.1227/NEU.0b013e318208f1da PMID: 21221039.
9. Lee M, Zaharchuk G, Guzman R, Achrol A, Bell-Stephens T, Steinberg GK. Quantitative hemodynamic studies in moyamoya disease: a review. Neurosurgical focus. 2009; 26(4):E5. doi: 10.3171/2009.1.FOCUS08300 PMID: 19335131; PubMed Central PMCID: PMC2905646.
10. Yun TJ, Cheon JE, Na DG, Kim WS, Kim IO, Chang KH, et al. Childhood moyamoya disease: quantitative evaluation of perfusion MR imaging—correlation with clinical outcome after revascularization surgery. Radiology. 2009; 251(1):216–23. doi: 10.1148/radiol.2511080654 PMID: 19332853.
11. Sugino T, Mikami T, Miyata K, Suzuki K, Houkin K, Mikuni N. Arterial spin-labeling magnetic resonance imaging after revascularization of moyamoya disease. Journal of stroke and cerebrovascular diseases: the official journal of National Stroke Association. 2013; 22(6):811–6. doi: 10.1016/j.jstrokecerebrovasdis.2012.05.010 PMID: 22721824.
12. Zhang J, Wang J, Geng D, Li Y, Song D, Gu Y. Whole-brain CT perfusion and CT angiography assessment of Moyamoya disease before and after surgical revascularization: preliminary study with 256-slice CT. PloS one. 2013; 8(2):e57595. doi: 10.1371/journal.pone.0057595 PMID: 23451248; PubMed Central PMCID: PMC3579776.
13. Uchino H, Kuroda S, Hirata K, Shiga T, Houkin K, Tamaki N. Predictors and clinical features of postoperative hyperperfusion after surgical revascularization for moyamoya disease: a serial single photon emission CT/positron emission tomography study. Stroke; a journal of cerebral circulation. 2012; 43(10):2610–6. doi: 10.1161/STROKEAHA.112.654723 PMID: 22871684.
14. Lee HY, Paeng JC, Lee DS, Lee JS, Oh CW, Cho MJ, et al. Efficacy assessment of cerebral arterial bypass surgery using statistical parametric mapping and probabilistic brain atlas on basal/

acetazolamide brain perfusion SPECT. Journal of nuclear medicine: official publication, Society of Nuclear Medicine. 2004; 45(2):202–6. PMID: 14960636.

15. Ashburner J. A fast diffeomorphic image registration algorithm. NeuroImage. 2007; 38(1):95–113. doi: 10.1016/j.neuroimage.2007.07.007 PMID: 17761438.

16. Fukui M. Guidelines for the diagnosis and treatment of spontaneous occlusion of the circle of Willis ('moyamoya' disease). Research Committee on Spontaneous Occlusion of the Circle of Willis (Moyamoya Disease) of the Ministry of Health and Welfare, Japan. Clin Neurol Neurosurg. 1997; 99 Suppl 2: S238–40. PMID: 9409446.

17. Research Committee on the P, Treatment of Spontaneous Occlusion of the Circle of W, Health Labour Sciences Research Grant for Research on Measures for Infractable D. Guidelines for diagnosis and treatment of moyamoya disease (spontaneous occlusion of the circle of Willis). Neurologia medico-chirurgica. 2012; 52(5):245–66. PMID: 22870528.

18. Kelly ME, Bell-Stephens TE, Marks MP, Do HM, Steinberg GK. Progression of unilateral moyamoya disease: A clinical series. Cerebrovascular diseases. 2006; 22(2–3):109–15. doi: 10.1159/000093238 PMID: 16685122.

19. Funaki T, Takahashi JC, Takagi Y, Yoshida K, Araki Y, Kikuchi T, et al. Incidence of late cerebrovascular events after direct bypass among children with moyamoya disease: a descriptive longitudinal study at a single center. Acta neurochirurgica. 2014; 156(3):551–9. doi: 10.1007/s00701-013-1975-7 PMID: 24363147.

20. Karasawa J, Touho H, Ohnishi H, Miyamoto S, Kikuchi H. Long-term follow-up study after extracranial-intracranial bypass surgery for anterior circulation ischemia in childhood moyamoya disease. J Neurosurg. 1992; 77(1):84–9. doi: 10.3171/jns.1992.77.1.0084 PMID: 1607976.

21. Ashburner J, Friston KJ. Unified segmentation. NeuroImage. 2005; 26(3):839–51. doi: 10.1016/j.neuroimage.2005.02.018 PMID: 15955494.

22. Takeuchi R, Yonekura Y, Takeda SK, Fujita K, Konishi J. Fully automated quantification of regional cerebral blood flow with three-dimensional stereotaxic region of interest template: validation using magnetic resonance imaging—technical note. Neurologia medico-chirurgica. 2003; 43(3):153–62. PMID: 12699126.

23. van Mook WN, Rennenberg RJ, Schurink GW, van Oostenbrugge RJ, Mess WH, Hofman PA, et al. Cerebral hyperperfusion syndrome. Lancet neurology. 2005; 4(12):877–88. doi: 10.1016/S1474-4422(05)70251-9 PMID: 16297845.

24. Houkin K, Kamiyama H, Abe H, Takahashi A, Kuroda S. Surgical therapy for adult moyamoya disease. Can surgical revascularization prevent the recurrence of intracerebral hemorrhage? Stroke; a journal of cerebral circulation. 1996; 27(8):1342–6. PMID: 8711799.

25. Miyamoto S, Japan Adult Moyamoya Trial G. Study design for a prospective randomized trial of extracranial-intracranial bypass surgery for adults with moyamoya disease and hemorrhagic onset—the Japan Adult Moyamoya Trial Group. Neurologia medico-chirurgica. 2004; 44(4):218–9. PMID: 15185765.

26. Miyamoto S, Yoshimoto T, Hashimoto N, Okada Y, Tsuji I, Tominaga T, et al. Effects of extracranial-intracranial bypass for patients with hemorrhagic moyamoya disease: results of the Japan Adult Moyamoya Trial. Stroke; a journal of cerebral circulation. 2014; 45(5):1415–21. doi: 10.1161/STROKEAHA.113.004386 PMID: 24668203.

27. Shigemoto Y, Matsuda H, Kamiya K, Maikusa N, Nakata Y, Ito K, et al. In vivo evaluation of gray and white matter volume loss in the parkinsonian variant of multiple system atrophy using SPM8 plus DARTEL for VBM. NeuroImage Clinical. 2013; 2:491–6. doi: 10.1016/j.nicl.2013.03.017 PMID: 24179801; PubMed Central PMCID: PMC3777846.

Significance of the Hemorrhagic Site for Recurrent Bleeding
Prespecified Analysis in the Japan Adult Moyamoya Trial

Jun C. Takahashi, MD, PhD; Takeshi Funaki, MD, PhD; Kiyohiro Houkin, MD, PhD; Tooru Inoue, MD, PhD; Kuniaki Ogasawara, MD, PhD; Jyoji Nakagawara, MD, PhD; Satoshi Kuroda, MD, PhD; Keisuke Yamada, MD, PhD; Susumu Miyamoto, MD, PhD; on behalf of the JAM Trial Investigators*

Background and Purpose—The primary results of the Japan Adult Moyamoya Trial revealed the statistically marginal superiority of bypass surgery over medical treatment alone in preventing rebleeding in moyamoya disease. The purpose of this analysis is to test the prespecified subgroup hypothesis that the natural course and surgical effects vary depending on the hemorrhagic site at onset.

Methods—The hemorrhagic site, classified as either anterior or posterior, was the only stratifying variable for randomization. Statistical analyses were focused on the assessment of effect modification according to the hemorrhagic site and were based on tests of interaction.

Results—Of 42 surgically treated patients, 24 were classified as anterior hemorrhage and 18 as posterior hemorrhage; of 38 medically treated patients, 21 were classified as anterior and 17 as posterior. The hazard ratio of the primary end points (all adverse events) for the surgical group relative to the nonsurgical group was 0.07 (95% confidence interval, 0.01–0.55) for the posterior group, as compared with 1.62 (95% confidence interval, 0.39–6.79) for the anterior group (P=0.013 for interaction). Analysis within the nonsurgical group revealed that the incidence of the primary end point was significantly higher in the posterior group than in the anterior group (17.1% per year versus 3.0% per year; hazard ratio, 5.83; 95% confidence interval, 1.60–21.27).

Conclusions—Careful interpretation of the results suggests that patients with posterior hemorrhage are at higher risk of rebleeding and accrue greater benefit from surgery, subject to verification in further studies.

Clinical Trial Registration—URL: http://www.umin.ac.jp/ctr/index.htm. Unique identifier: C000000166.

(*Stroke*. 2016;47:37-43. DOI: 10.1161/STROKEAHA.115.010819.)

Key Words: cerebral revascularization ■ confidence intervals ■ incidence ■ intracerebral hemorrhage ■ Japan ■ moyamoya disease

In adult-onset moyamoya disease, intracranial hemorrhage accounts for one half of primary manifestations. The rebleeding rate (bleeding rate after initial bleeding) is ≈7% per year, and the outcome after rebleeding is poor.[1] The Japan Adult Moyamoya (JAM) Trial, a multicenter randomized controlled trial, was conducted to determine whether direct bypass surgery reduces rebleeding in adult-onset hemorrhagic moyamoya disease. The published primary results support this hypothesis.[2]

Hemorrhage can result from the various fragile collaterals specific to moyamoya disease, including tiny collateral vessels that develop from the lenticulostriate arteries and the dilated abnormal branches from the thalamic and choroidal arteries.[3,4] This diversity of vessels contributes to the potential for hemorrhage in several regions: the basal ganglia, thalamus, intra- or periventricular region, and subarachnoid space.[5] If the benefit of bypass surgery in preventing rebleeding is explained as elimination of hemodynamic stress in fragile collaterals,[6,7] the surgical effects will vary with the bleeding site because the surgery targets a specific territory. Rebleeding risk with nonsurgical treatment might also be heterogeneous in the relation to the bleeding site. In light of these hypotheses, randomization in the JAM Trial was stratified according to

Received July 14, 2015; final revision received October 21, 2015; accepted November 3, 2015.

From the Department of Neurosurgery (J.C.T.) and Integrative Stroke Imaging Center (J.N.), National Cerebral and Cardiovascular Center, Suita, Japan; Department of Neurosurgery, Kyoto University Graduate School of Medicine, Kyoto, Japan (T.F., K.Y., S.M.); Department of Neurosurgery, Hokkaido University Graduate School of Medicine, Sapporo, Japan (K.H.); Department of Neurosurgery, Faculty of Medicine, Fukuoka University, Fukuoka, Japan (T.I.); Department of Neurosurgery, Iwate Medical University, Morioka, Japan (K.O.); and Department of Neurosurgery, Graduate School of Medicine and Pharmaceutical Sciences, University of Toyama, Toyama, Japan (S.K.).

*A list of all JAM Trial Investigators is given in the Appendix.

The online-only Data Supplement is available with this article at http://stroke.ahajournals.org/lookup/suppl/doi:10.1161/STROKEAHA.115.010819/-/DC1.

Correspondence to Susumu Miyamoto, MD, PhD, Department of Neurosurgery, Graduate School of Medicine, Kyoto University, 54 Shogoin Kawahara-cho, Sakyo-ku, Kyoto, 606-8507, Japan. E-mail miy@kuhp.kyoto-u.ac.jp

***Stroke* is available at http://stroke.ahajournals.org** **DOI: 10.1161/STROKEAHA.115.010819**

bleeding site at onset through classification as either anterior or posterior determined before randomization.

This subgroup analysis is intended to test the above prespecified hypothesis about hemorrhagic moyamoya disease: that the anterior and posterior hemorrhage groups differ in terms of the outcome of nonsurgical treatment and the effect of bypass surgery at preventing rebleeding.

Methods

The design of the JAM Trial has been described elsewhere.[2,8] In brief, participants were recruited from 22 centers specializing in moyamoya disease. Patients with moyamoya disease were eligible if they had suffered intracranial hemorrhage within the 12 months before randomization, were aged 16 to 65 years, were independent in daily life (0–2 for modified Rankin Scale score), had completed acute phase treatment at least 1 month before randomization, and had been free from ischemic/hemorrhagic attack for at least 1 month. Diagnoses of moyamoya disease were made according to the proposed criteria.[9] Participants were randomly allocated to either the surgical or the nonsurgical group. Stratified randomization according to hemorrhagic site was adopted, as described later. Those in the surgical group required direct bypass surgery, including superficial temporal artery–middle cerebral artery anastomosis, within 3 months of randomization in addition to medical treatment, and those in the nonsurgical group underwent medical treatment alone. The primary end points were defined as any of the following events: a rebleeding attack, completed stroke resulting in significant morbidity, significant morbidity or mortality from any medical cause, or requirement for bypass surgery for a nonsurgical patient as determined by a registered neurologist. The secondary end point was defined as a rebleeding attack. Both a neurologist and a neurosurgeon followed each participant in each participating institute for 5 years or until end points were reached. Although antihypertensive medication was administrated to patients with hypertension, the use of anticoagulants or antiplatelet drugs was prohibited. The patients' medical, neurological, radiological, and functional conditions were closely monitored and reported annually. The study was approved by the ethical committees of all participating centers and was registered with the University Hospital Medical Information Network Clinical Trials Registry (UMIN-CTR, ID: C000000166, 2005).

Subgroup of Interest

Hemorrhagic site was the only stratifying variable for randomization. The study protocol allowed for analyses of the subgroup in advance. Before randomization, the study office classified the hemorrhage at onset in each participant as either anterior or posterior according to the center location of the intracerebral hemorrhage as observed in computed tomographic images (Figure 1; Figures I–IV in the online-only Data Supplement). An anterior hemorrhage was defined as one attributable to perforating arteries from the anterior or middle cerebral artery, including those located in the putamen, caudate head, frontal lobe, anterior half of the temporal lobe, subependymal area of the anterior part of the lateral ventricle, or anterior half of the corpus callosum. A posterior hemorrhage was defined as one attributable to perforating arteries from the posterior cerebral artery or choroidal arteries, including those located in the thalamus, posterior half of the temporal lobe, parietal lobe, occipital lobe, subependymal area of the posterior part of the lateral ventricle including the trigon, or posterior half of the corpus callosum. Primary intraventricular hemorrhage, defined as intraventricular hemorrhage without intraparenchymal hemorrhage, was classified as either anterior or posterior according to the distribution of hematoma (Figure 1A). Any diffusely distributed primary intraventricular hemorrhage whose origin was difficult to determine was classified as anterior. Subarachnoid hemorrhage without intracerebral hemorrhage was classified in a similar fashion (Figure 1A).

Statistical Analysis

All analyses were performed according to the intention-to-treat principle. Kaplan–Meier survival analysis and Cox proportional hazard model were used to compare the incidence of the end points in each subgroup. The assessment of effect modification (heterogeneity of

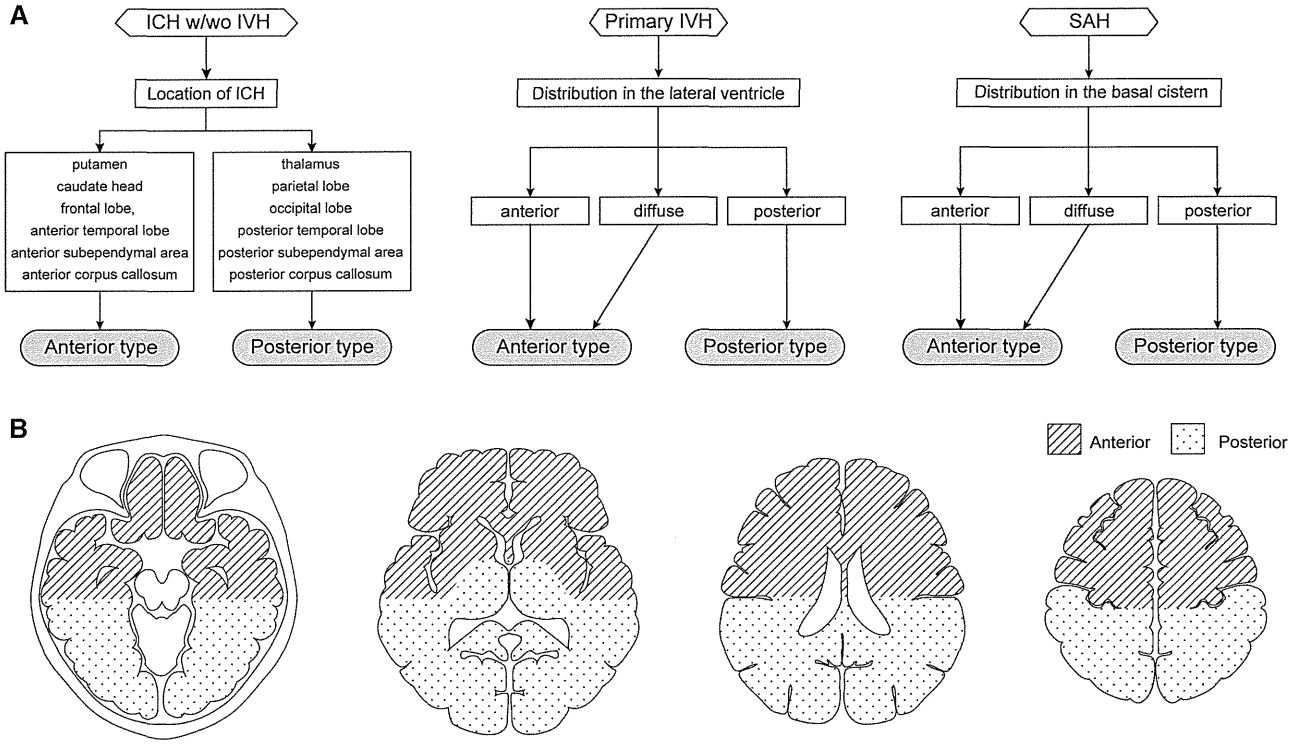

Figure 1. A, Flow diagram showing the algorithm for classifying hemorrhagic sites. **B**, Schematic illustrations showing topographical definitions of hemorrhagic sites. ICH indicates intracerebral hemorrhage; IVH, intraventricular hemorrhage; and SAH, subarachnoid hemorrhage.