◀ **Fig. 1** Intraoperative photographs show the step-by-step procedures of superficial temporal artery to middle cerebral artery (STA-MCA) anastomosis and encephalo-duro-myo-arterio-pericranial synangiosis (EDMAPS). **a** Following the curved skin incision, the frontal (*) and parietal branches of the STA (**) are dissected under a surgical microscope. **b** The temporal muscle (*M*) and frontal pericranium (*P*) are dissected from the skull and preserved as the vascularized flaps. **c** The burr hole just rostral to the pterion (*arrow*) is made to locate the anterior branch of the middle meningeal artery (*MMA*). **d** Heart-shaped frontotemporal craniotomy is performed, leaving the lesser wing of the sphenoid bone intact (*arrow*). Note the wide extension of the craniotomy to the frontal area. **e** The lesser wing of the sphenoid bone is carefully resected with a rongeur or high-speed diamond drill to preserve the anterior branch of the MMA (*arrow*). **f** The dura mater is opened, keeping the main branches of the MMA intact. The frontal branch of the STA is anastomosed to the cortical branch of the MCA. The dural pedicles are turned into the epiarachnoid space. **g** The dural windows are closed with the temporal muscle (*M*) and frontal pericranium (*P*)

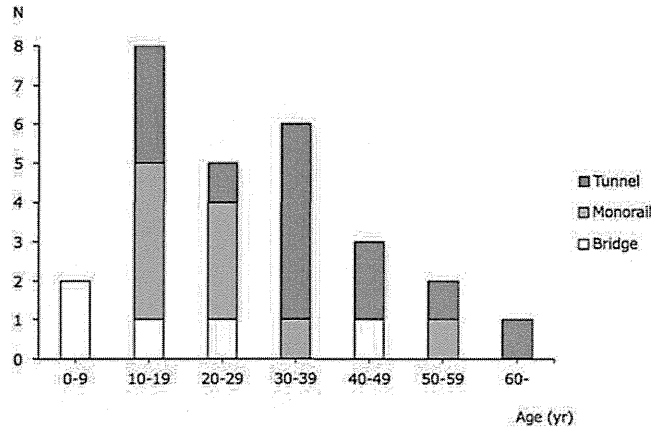

Fig. 3 Column graph shows the relationship between patient age and anatomical findings of the anterior branch of the MMA around the pterion

groups: the patients younger than 30 years and those older (χ^2 test, p=0.0168).

Table 2 summarizes the relationship between the anatomy of the MMA branch and its preservation during craniotomy. It was quite easy to preserve the bridge-type anterior branch of the MMA by piecemeal resection of the lesser wing of the sphenoid bone with a rongeur. The bridge-type MMA branch could be preserved during surgery in all five sides (Fig. 4a–c). Careful piece-by-piece resection of the lesser wing with a rongeur was essential to preserve monorail-type MMAs, because the MMA branch was tightly attached to the cranium inside the groove. As a result, the monorail-type MMA branch could be preserved in five (71.4 %) of seven sides when a rongeur was used to remove the lesser wing. However, it was quite easy to preserve the monorail-type MMA branch in all three sides (100 %) when the lesser wing was carefully drilled out using a high-speed diamond drill. On the other hand, the tunnel-type MMA branches could not be preserved in spite of careful bone resection with a rongeur because they were tightly attached to the entirely surrounding bone. Therefore, the lesser wing was

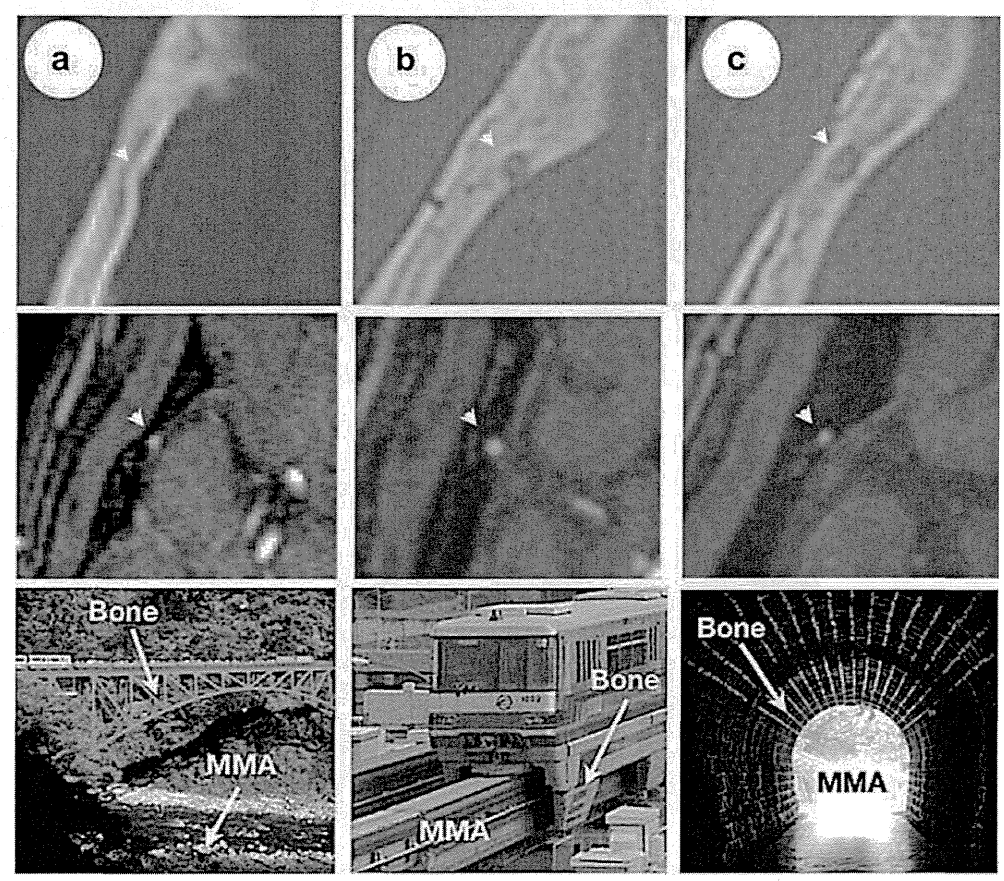

Fig. 2 The anatomical relationship between the anterior branch of the MMA and lesser wing of the sphenoid bone is classified into the bridge (**a**), monorail (**b**), or tunnel type (**c**). Upper panels demonstrate the representative findings of the plain CT scan in each type. Arrows show a shallow groove (**a**), deep groove (**b**), and bony tunnel (**c**). The middle panels show the representative findings of raw images on time-of-flight MR angiography in each type. Arrows indicate the flow signal of the anterior branch of the MMA. Lower panels show figurative photographs of each type

Table 2 The rate of preservation of the MMA using a high-speed drill

Type	N (%)	Drill	Preservation (%)
Bridge	5 (18.5)	−	5 (100)
Monorail	7 (25.9)	−	5 (71.4)
	3 (11.1)	+	3 (100)
Tunnel	5 (18.5)	−	0 (0.0)
	7 (25.9)	+	7 (100)

drilled out to preserve tunnel-type MMAs using a high-speed diamond drill, which made it possible to preserve it in all seven sides (Fig. 4d–f). The patency of the anterior branch of the MMA could easily be confirmed using intraoperative ICG videoangiography. Postoperative cerebral angiography and/or MR angiography could also evaluate it in all patients (Fig. 5).

The results show that no ischemic stroke occurred within 7 days after surgery. In addition, no patients experienced any ischemic or hemorrhagic stroke during a mean follow-up period of 16 months.

Discussion

This study clearly demonstrates a significant anatomical variation of the anterior branch of the MMA around the pterion among patients with moyamoya disease. The anterior branch of the MMA ran just within the middle meningeal groove in 15 (55.6 %) of 27 sides. The groove was quite shallow in 5/15 (bridge type), but deep in 10/15 (monorail type). On the other hand, the anterior branch of the MMA penetrated the bony tunnel in the lesser wing of the sphenoid bone in the other 12 (44.4 %) of 27 sides. The finding differs greatly from those in previous studies. For example, Shimizu et al. (2008) found 59 tunnels on 78 sides (75.6 %). Ma et al. (2012) also reported that the vascular marking for the anterior branch of the MMA consisted of a groove in 30 % of 152 sides and a tunnel in the other 70 %. Age differences of the samples in each study may explain this discrepancy. Thus, these previous studies analyzed only adult skulls. However, the present study included 9 children and 13 adults with moyamoya disease. In fact, statistical analysis reveals a significant difference in the anatomical relationship between patients younger than 30 years and those older. Therefore, the bony tunnel may develop with growth, although there are no previous studies on this issue.

From the viewpoint of surgical revascularization for moyamoya disease, this study provides novel information on developing a surgical technique to preserve the anterior branch of the MMA, which can provide important collateral flow to the ACA territory. As reported previously, a heart-shaped craniotomy with a burr hole rostral to the pterion can make it easy to preserve the anterior branch of the MMA during craniotomy in all patients with moyamoya disease [4, 9]. As the next step, in order to preserve the anterior branch of the MMA when resecting the lesser wing of the sphenoid, it is essential to recognize the anatomical relationship between them. In this study, based on our experience, their relationships were classified into three types: the bridge, monorail, and tunnel. In all patients with bridge-type MMA, the resection of the lesser wing with a ronguer preserved it quite easily. In a majority of patients with monorail-type MMA, careful and piecemeal resection with a rongeur could preserve it, although it was sometimes impossible, especially when the middle meningeal groove was very deep. In this study, it was difficult to preserve tunnel-type

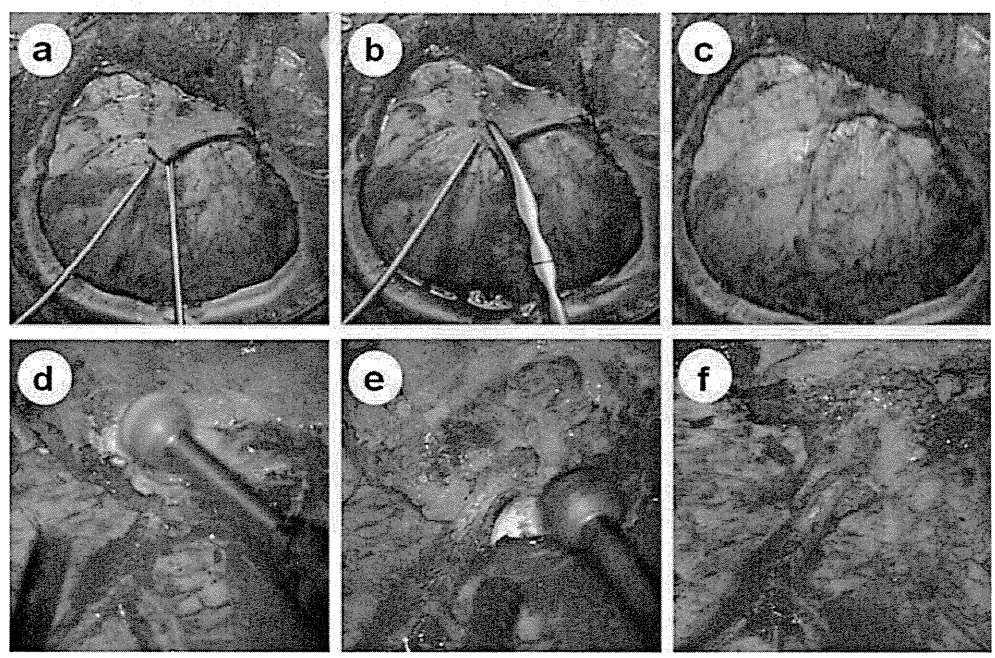

Fig. 4 Intraoperative photographs demonstrate surgical techniques to resect the lesser wing of the sphenoid bone after heart-shaped frontotemporal craniotomy with a rongeur (**a-c**) or high-speed diamond drill (**d-f**)

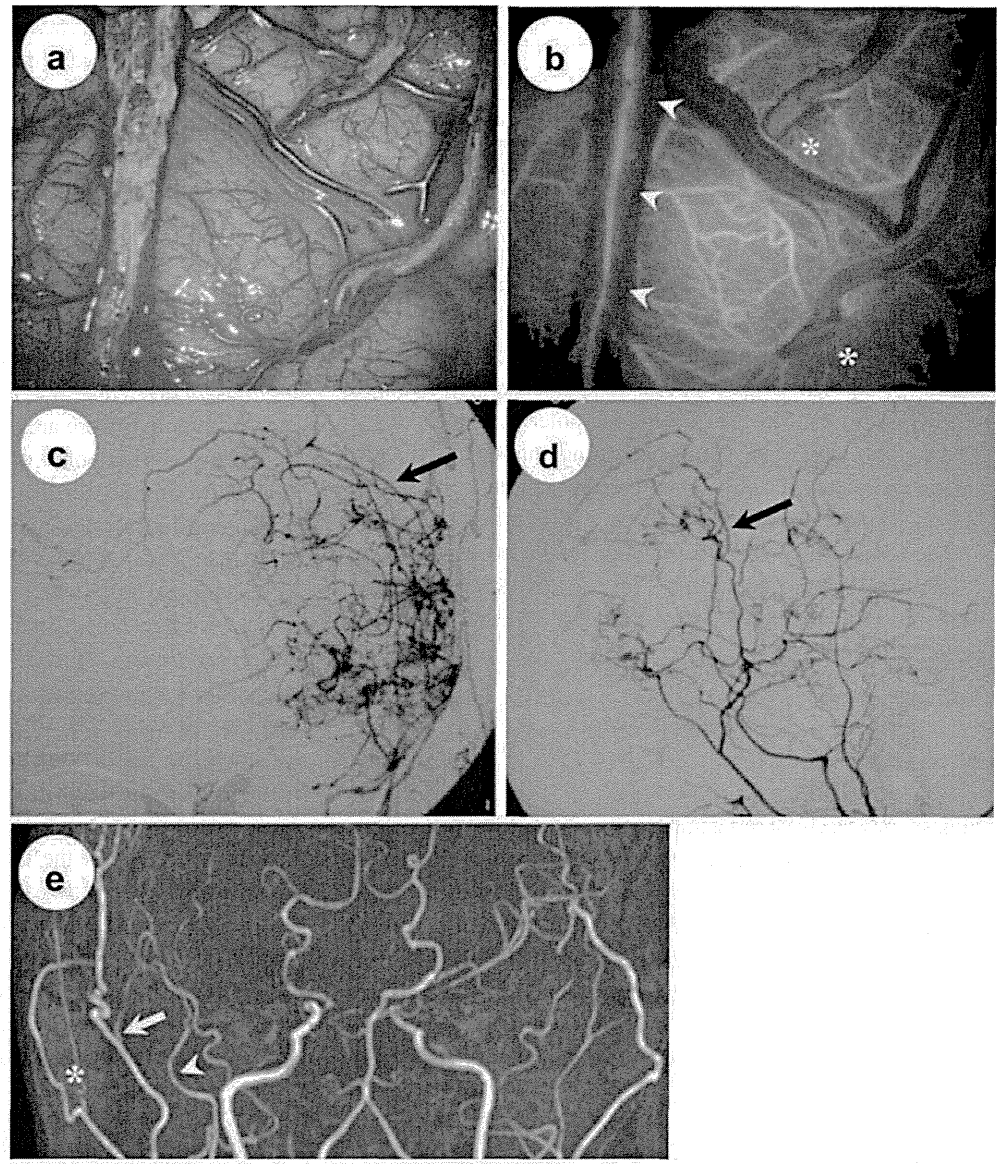

Fig. 5 White-light (**a**) and near infrared photographs (**b**) during surgery reveal that intraoperative ICG videoangiography can visualize the blood flow in the preserved MMA (*arrowheads*) as well as the STA-MCA anastomosis (*). Anteroposterior (**c**) and lateral (**d**) projection of postoperative external carotid angiography demonstrates that the anterior branch of the MMA is preserved and supplies blood flow to the anterior cerebral artery territory (*arrows*). Postoperative MR angiography (**e**) reveals that the STA (*), MMA (*arrowhead*), and the deep temporal artery (*arrow*) are patent 4 months after surgery

MMA when resecting the lesser wing with a rongeur, mainly because of the long (~15 mm) bony tunnel, despite the small caliber of the MMA [10]. On the other hand, it was possible to preserve it when resecting with a high-speed diamond drill under a surgical microscope. The resection of the lesser wing of the sphenoid is essential to performing STA-MCA double anastomosis targeted to the frontal and temporal branches of the MCA and to yield a wide attachment between the temporal lobe and temporal muscle, an important donor tissue for an indirect bypass. The clinical significance of the surgical technique in this study is still obscure. However, it is valuable to maintain the cerebral hemodynamics in the frontal lobe, especially in pediatric patients, because they are at higher risk for poor intellectual outcome [15]. Longer follow-up study in a larger cohort is warranted to evaluate this hypothesis.

In this study, the anatomical relationship between the anterior branch of the MMA and lesser wing of the sphenoid bone could be evaluated using MR angiography or 3D-CTA. These less invasive modalities allowed us to anticipate it before surgery. In addition, ICG videoangiography was quite useful to evaluate whether the anterior branch of the MMA could be preserved or not during surgery. The findings on ICG videoangiography correlated very well with postoperative cerebral angiography and/or MR angiography. MR angiography can easily visualize the increased calibers of the branches of the external carotid artery that participate in surgical collaterals. The branches include the STA, MMA, and deep temporal artery (DTA), which normally supply the blood flow to the temporal muscle [16]. In this study, postoperative radiological examinations were performed 3 to 4 months after surgery. The results showed that MR angiography clearly revealed all of these branches increased their calibers by providing collateral blood flow through the indirect bypass after surgery as well as confirmed the patency of the anastomosis.

Conclusion

It is essential to understand the surgical anatomy of the MMA around the pterion in order to preserve its anterior branch during surgery because it can potentially function as important collaterals to the ACA territory in moyamoya disease. Bridge and monorail-type MMA can be preserved by carefully resecting the lesser wing of the sphenoid bone. However, the lesser wing should be drilled out with a high-speed diamond drill under a surgical microscope to preserve the tunnel-type MMA. Pre-, intra-, and postoperative imaging modalities such as MR angiography, 3D-CTA, ICG videoangiography, and cerebral angiography are quite useful to evaluate the surgical anatomy and preservation of MMA during bypass surgery for moyamoya disease.

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Conflict of interest None

References

1. Kuroda S, Houkin K (2008) Moyamoya disease: current concepts and future perspectives. Lancet Neurol 7:1056–1066
2. Suzuki J, Takaku A (1969) Cerebrovascular "moyamoya" disease. Disease showing abnormal net-like vessels in base of brain. Arch Neurol 20:288–299
3. 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
4. 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
5. Matsushima T, Inoue K, Kawashima M, Inoue T (2012) History of the development of surgical treatments for moyamoya disease. Neurol Med Chir (Tokyo) 52:278–286
6. 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
7. 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
8. Dusick JR, Gonzalez NR, Martin NA (2011) Clinical and angiographic outcomes from indirect revascularization surgery for moyamoya disease in adults and children: a review of 63 procedures. Neurosurgery 68:34–43, discussion 43
9. Kuroda S, Houkin K (2012) Bypass surgery for moyamoya disease—concept and essence of surgical technique. Neurol Med Chir (Tokyo) 52:287–294
10. Harthmann da Silva T, Ellwanger JH, Silva HT, Moraes D, Dotto AC, Viera Vde A, de Campos D (2013) Morphometric analysis of the middle meningeal artery organization in humans—embryological considerations. J Neurol Surg Part B, Skull base 74: 108–112
11. Plummer SC III (1896) Research on the surgical anatomy of the middle meningeal artery. Ann Surg 23:540–572
12. Ma S, Baillie LJ, Stringer MD (2012) Reappraising the surface anatomy of the pterion and its relationship to the middle meningeal artery. Clin Anat 25:330–339
13. Shimizu S, Hagiwara H, Utsuki S, Oka H, Nakayama K, Fujii K (2008) Bony tunnel formation in the middle meningeal groove: an anatomic study for safer pterional craniotomy. Minim Invasive Neurosurg 51:329–332
14. Research Committee on the Pathology and Treatment of Spontaneous Occlusion of the Circle of Willis (2012) Guidelines for diagnosis and treatment of moyamoya disease (spontaneous occlusion of the circle of Willis). Neurol Med Chir (Tokyo) 52:245–266
15. Kuroda S, Houkin K, Ishikawa T, Nakayama N, Ikeda J, Ishii N, Kamiyama H, Iwasaki Y (2004) Determinants of intellectual outcome after surgical revascularization in pediatric moyamoya disease: a multivariate analysis. Childs Nerv Syst 20:302–308
16. Houkin K, Nakayama N, Kuroda S, Ishikawa T, Nonaka T (2004) How does angiogenesis develop in pediatric moyamoya disease after surgery? A prospective study with MR angiography. Childs Nerv Syst 20:734–741

Comments

This is an important article about preservation of the MMA frontal branch at the time of craniotomy during the STA-MCA bypass surgery for Moyamoya disease. The meaning of this preservation is to protect the collateral circulation to the anterior cerebral artery ACA via the connecting vasculature of the Falx cerebri, as the paper indicates. The perfusion territory of the ACA is considered to be important for the intellectual development and its maintenance. On the other hand, direct STA-ACA bypass would come into question although this topic has not been discussed in the article : How to perform the STA-ACA bypass? , preservation of the MMA towards the midline at the time of craniotomy for the bypass, and so on. With the addition of the latter the article would have been more interesting and exciting.

Y.Yonekawa
Kyoto, Japan

Reference:
Yonekawa Y (2009) Brain revascularization by extracranial-intracranial arterial bypass In Sindou M (ed.): Practical Handbook of Neurosurgery, Springer, Vol 1 pp355-381

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Asymptomatic Moyamoya Disease: Literature Review and Ongoing AMORE Study

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Abstract

Recent development of a non-invasive magnetic resonance examination has increased the opportunity to identify asymptomatic patients with moyamoya disease who have experienced no cerebrovascular events. However, their clinical features, prognosis, and treatment strategy are still unclear because of small number of subjects and short follow-up periods. Therefore, we have designed Asymptomatic Moyamoya Registry (AMORE) study in Japan. The objectives of this nation-wide, multi-center prospective study are to clarify long-term prognosis of asymptomatic patients with moyamoya disease and to determine the risk factors that cause ischemic and hemorrhagic stroke in them. In this article, we review the published data on asymptomatic moyamoya disease and report the on-going multi-center prospective cohort study, AMORE study. We would like to emphasize the importance to determine the clinical features, prognosis, and treatment strategies of asymptomatic moyamoya disease in very near future.

Key words: asymptomatic moyamoya disease, AMORE study, prognosis

Introduction

Moyamoya disease is a unique cerebrovascular disorder characterized by progressive stenosis of the terminal portion of the internal carotid artery (ICA). The perforating arteries in the basal ganglia and thalamus markedly dilate and function as an important collateral circulation, called as "moyamoya" vessels. The posterior cerebral arteries are also involved in a certain subgroup of patients. Therefore, cerebral hemodynamics is often impaired especially in the frontal lobe, leading to transient ischemic attack (TIA) and cerebral infarction. Furthermore, the dilated, fragile moyamoya vessels often rupture and cause intracranial hemorrhage.[1,2] The etiology of the disease is still unknown; however, recent studies have strongly suggested the involvement of some genetic factors in its pathogenesis.[3] The potential contribution of infections has also been pointed out, although specific pathogens have not been identified. Superficial temporal artery to middle cerebral artery (STA-MCA) anastomosis and indirect synangiosis are well known to improve cerebral hemodynamics and reduce the risk of subsequent cerebrovascular events, including both ischemic and hemorrhagic stroke, and improve long-term outcome in patients with moyamoya disease.[4–11]

On the other hand, the recent development of noninvasive diagnostic modalities, including magnetic resonance imaging (MRI) and magnetic resonance angiography (MRA), has shown that the incidence of asymptomatic moyamoya disease may be much higher than previously believed (*see below*). Even in Japan, however, the epidemiology of asymptomatic moyamoya disease is still obscure, and the guidelines for the management of asymptomatic moyamoya disease have not yet been established. In this article, we review the published data on asymptomatic moyamoya disease and report the on-going multi-center prospective cohort study, Asymptomatic Moyamoya Registry (AMORE) study.

Definition

All patients should meet the guidelines for the diagnosis of moyamoya disease set by the Research Committee on Moyamoya Disease of the Ministry of Health, Labor, and Welfare of Japan. All patients should previously have no ischemic or hemorrhagic episode and be neurologically free. Patients who had previously experienced any episode suggestive

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of TIA, cerebral infarction, intracranial hemorrhage, seizure, or involuntary movement caused by moyamoya disease should be excluded. Careful medical interview should be performed to distinguish moyamoya disease-related headache from non-specific headache such as tension-type headache.[12]

Epidemiology

The incidence of moyamoya disease is high in countries in East Asia such as Japan and Korea. In Japan, the annual prevalence has been estimated to be 3.16–10.5 per 100,000. The female to male ratio was shown to be 1.8.[2] However, both the prevalence and incidence of asymptomatic moyamoya disease are still unclear. Previously, asymptomatic patients with moyamoya disease have rarely been sporadically reported. Screening of family members with moyamoya disease has also identified small number of asymptomatic patients. Therefore, the incidence of asymptomatic moyamoya disease had been believed very low. In fact, Yamada et al. (2005) reported the results of a nation-wide questionnaire conducted in 1994 and identified 33 asymptomatic patients (1.5%) out of a total of 2,193 patients.[13] Nanba et al. (2003) reviewed their single-center experiences and precisely reported the clinical features of 10 asymptomatic patients with moyamoya disease.[14] Furthermore, an all-inclusive survey of moyamoya disease in Hokkaido, one of the major islands in Japan, revealed that 47 (17.8%) of 267 newly enrolled patients between 2002 and 2006 were asymptomatic.[15]

The first multi-center, nation-wide survey focused on asymptomatic patients with moyamoya disease was conducted between 2003 and 2006 in Japan. As a result, totally 40 patients were enrolled from 12 hospitals. Their mean age was 41.4 years ranging from 13 years to 67 years. The female-to-male ratio was 2.1. Clues to the diagnosis were tension-type headache in 14 patients, dizziness in 5, and head trauma in 4. Five patients were incidentally diagnosed on MRI and MRA performed for a brain health check-up. Five diagnoses were made on MRI and MRA performed for screening, because a member of their family had moyamoya disease diagnosed. They were siblings in two and offspring in three. The remaining seven cases were diagnosed on MRI and MRA performed because of an unrelated disease in other organs. Therefore, the prevalence and incidence may be much higher than considered before. The female-to-male ratio and mean age of the patients in these studies were very similar to those of moyamoya disease as a whole.[12]

Radiological Findings

On cerebral angiography, the bilateral carotid forks were involved in a majority of asymptomatic patients with moyamoya disease. Thus, Nanba et al. (2003) reported that all 10 patients were classified into bilateral type.[14] Subsequently multi-center, nation-wide survey in Japan also reported that 37 of 40 asymptomatic patients were judged as bilateral type. Of 72 involved hemispheres, 33 (45.8%) and 21 (29.2%) were graded as stage 3 and stage 4, respectively. More importantly, older patients had significantly more advanced disease stage ($P = 0.0134$).[12]

Cerebral infarction was identified in a certain subgroup of asymptomatic patients with moyamoya disease, although they experienced no cerebrovascular events. Nanba et al. (2003) reported that 3 (30%) of 10 asymptomatic patients had cerebral infarction in the watershed zone.[14] Multi-center, nation-wide survey in Japan also reported that cerebral infarction was identified in 16 (20.8%) of 77 involved hemispheres.[12] According to a population-based autopsy study in Japan, the incidence of silent cerebral infarction was 4.4% in 40–59-year-old people.[16] Therefore, the incidence of silent cerebral infarction is much higher in asymptomatic moyamoya disease than in normal population.

None of these studies detected intracranial hemorrhage in asymptomatic moyamoya disease. However, recent studies have demonstrated that T_2*- or susceptibility-weighted MRI can more sensitively detect silent microbleeds in moyamoya disease than conventional MRI. Thus, 15–44% of adult patients with moyamoya disease have silent microbleeds in the basal ganglia, thalamus, and periventricular white matter, where they are prone to intracranial hemorrhage. Silent microbleeds may also be an independent predictor for subsequent hemorrhagic stroke.[17–21] More notably, Kuroda et al. (2013) reported that silent microbleeds were even found in 5 (25%) of 20 asymptomatic patients.[21] Therefore, further study would be warranted to evaluate the incidence of silent microbleeds in larger cohort of asymptomatic moyamoya disease.

The data on cerebral hemodynamics is limited in asymptomatic patients with moyamoya disease. Thus, Nanba et al. (2003) quantified cerebral blood flow (CBF) and cerebrovascular reactivity (CVR) to acetazolamide in 10 asymptomatic patients and found that 2 patients had normal CBF but reduced CVR and other two had reduced CBF and CVR in the involved middle cerebral artery (MCA) territory.[14] Multi-center, nation-wide survey in Japan

also reported that 24 (34.3%) of 70 involved hemispheres had normal CBF but reduced CVR and other 7 (10%) had reduced CBF and CVR. Therefore, cerebral hemodynamics may be disturbed even in about 40% of asymptomatic patients.[12] The findings are quite important to further consider the prognosis and treatment strategy in asymptomatic moyamoya disease, because the patients with reduced CBF and CVR due to occlusive carotid artery diseases are known to be at higher risk for subsequent ischemic stroke.

Prognosis

As aforementioned, the long-term prognosis in asymptomatic patients with moyamoya disease is not fully understood. Based on a nation-wide questionnaire study conducted in 1994, Yamada et al. (2005) retrospectively analyzed the prognosis in 33 asymptomatic patients and found that 4 patients developed TIA and other two died of intracranial hemorrhage.[13] Nanba et al. (2003) followed up 10 asymptomatic patients during a mean period of 4.1 years. As a result, one patient (10%) developed ischemic stroke due to the progression of disease stage.[14]

Multi-center, nation-wide survey in Japan was the first historical prospective cohort study to evaluate the prognosis in asymptomatic patients. Of totally 40 patients enrolled in this study, 6 underwent surgical revascularization including STA-MCA anastomosis, and other 34 were conservatively followed up. Of these, antiplatelets and/or anticonvulsants were prescribed in 11 asymptomatic patients. During a mean period of 43.7 months, 6 surgically treated patients experienced no cerebrovascular events. On the other hand, 7 of 34 conservatively treated patients developed any cerebrovascular events, including TIA in three patients, ischemic stroke in one, and hemorrhagic stroke in three. As a result, the annual risk of any cerebrovascular events and stroke was 5.7% and 3.2%, respectively. Disturbed cerebral hemodynamics at initial diagnosis was significantly linked to ischemic episodes ($P < 0.05$). Disease progression during follow-up periods also highly caused ischemic episodes. Follow-up MRI and MRA revealed silent radiological changes in other three patients, including cerebral infarction, microbleed, and disease progression.[12] As previously reported, disease progression occurs in about 20% of patients during a mean follow-up period of 6 years. Occlusive arterial lesions progress in both anterior and posterior circulation, in both bilateral and unilateral types, and in both symptomatic and asymptomatic patients. Multivariate analysis has revealed that female gender is an independent risk factor for disease progression. Therefore, it would be natural that disease progression occurs and causes ischemic or hemorrhagic stroke even in asymptomatic patients.[22]

Based on these observations, asymptomatic moyamoya disease is not a "silent" disorder and readily progress to cause ischemic and hemorrhagic stroke. It would also be essential to repeat MRI and MRA at regular intervals when asymptomatic patients are conservatively followed up to detect disease progression before any cerebrovascular events occur.

Management

Surgical revascularization has widely been accepted to reduce the risk of subsequent ischemic and hemorrhagic stroke in symptomatic patients with moyamoya disease.[4–11] However, management strategies for asymptomatic moyamoya disease have not been established yet because of limited information on its clinical features.

The Research Committee on Moyamoya Disease in Japan recommends the management of risk factors and lifestyle guidance. Antiplatelet agents are not recommended for asymptomatic patients, because they may suffer hemorrhagic stroke. Surgical revascularization may be indicated, at least, in asymptomatic patients with disturbed cerebral hemodynamics, if surgical morbidity is low enough. As aforementioned, precise and regular MRI/MRA examinations should be repeated to improve their long-term outcome by predicting ischemic and hemorrhagic stroke before the onset.[23] However, the first multi-center, nation-wide survey in Japan was a historical prospective cohort study and not a prospective study.[12] Therefore, a prospective cohort or randomized study is warranted on the basis of a larger population of asymptomatic patients to build accurate evidence on the clinical features and outcome of asymptomatic moyamoya disease.

AMORE Study

Based on these observations, the Research Committee on Moyamoya Disease in Japan conducted a prospective multi-center, nation-wide observational study, AMORE study, in January 2012 to further clarify the epidemiology, pathophysiology, and prognosis in asymptomatic moyamoya disease. This study is done at 20 centers in Japan (*see Appendix*). The study confirmed to the Helsinki Declaration, and Good Clinical Practice Guideline, and was approved by the ethics committees at participating centers.

I. Patient eligibility

Patients are eligible if they meet the following criteria: age 20–70 years; bilateral or unilateral moyamoya disease on cerebral angiography or MRA; no episodes suggestive of TIA, ischemic stroke, and hemorrhagic stroke; possible to conservatively follow-up; and independent in daily life (modified Rankin scale 0 or 1). Exclusion criteria are previous episodes suggestive of TIA, ischemic stroke, and hemorrhagic stroke, and quasi-moyamoya disease. The patients are registered for 4 years between January 2012 and December 2015. All patients provide written informed consent when included in this study. Clinical information at enrollment includes patient's age, gender, clue of diagnosis, past history, family history of moyamoya disease, modified Rankin scale, medicine, the frequency, location, and severity of headache, laboratory data, blood pressure, MRI [T_2-weighted images, T_2*-weighted images, and fluid-attenuated inversion recovery (FLAIR) images], MRA (3-dimensional time-of-flight) or cerebral angiography, and single photon emission computed tomography/ positron emission tomography (SPECT/PET) data.

II. Follow-up

All enrolled patients are followed up for 5 years. A follow-up assessment is scheduled at 12 months, including any cerebrovascular event, blood pressure, MRI (T_2-weighted images, T_2*-weighted images, and FLAIR images), and MRA (3-dimensional time-of-flight). Primary endpoint is any ischemic and hemorrhagic stroke during a follow-up period of 5 years. Any ischemic stroke includes fresh cerebral infarction on diffusion-weighted MRI in spite of clinically transient neurological deficits that resolve within 24 hours after the onset. Secondary outcomes are TIA, newly developed ischemic and hemorrhagic lesions, and disease progression during a follow-up period of 5 years.

Conclusion

Clinical features and outcomes of asymptomatic moyamoya disease should be clarified by conducting further studies, including ongoing AMORE study. Treatment strategies would be established through these efforts.

Acknowledgments

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Conflicts of Interest Disclosure

None.

References

1) Suzuki J, Takaku A: Cerebrovascular "moyamoya" disease. Disease showing abnormal net-like vessels in base of brain. *Arch Neurol* 20: 288–299, 1969
2) Kuroda S, Houkin K: Moyamoya disease: current concepts and future perspectives. *Lancet Neurol* 7: 1056–1066, 2008
3) Liu W, Morito D, Takashima S, Mineharu Y, Kobayashi H, Hitomi T, Hashikata H, Matsuura N, Yamazaki S, Toyoda A, Kikuta K, Takagi Y, Harada KH, Fujiyama A, Herzig R, Krischek B, Zou L, Kim JE, Kitakaze M, Miyamoto S, Nagata K, Hashimoto N, Koizumi A: Identification of RNF213 as a susceptibility gene for moyamoya disease and its possible role in vascular development. *PLoS ONE* 6: e22542, 2011
4) 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* 77: 84–89, 1992
5) Kawaguchi S, Okuno S, Sakaki T: Effect of direct arterial bypass on the prevention of future stroke in patients with the hemorrhagic variety of moyamoya disease. *J Neurosurg* 93: 397–401, 2000
6) Kuroda S, Houkin K, Ishikawa T, Nakayama N, Iwasaki Y: Novel bypass surgery for moyamoya disease using pericranial flap: its impacts on cerebral hemodynamics and long-term outcome. *Neurosurgery* 66: 1093–1101; discussion 1101, 2010
7) Kuroda S, Houkin K, Kamiyama H, Abe H, Mitsumori K: Regional cerebral hemodynamics in childhood moyamoya disease. *Childs Nerv Syst* 11: 584–590, 1995
8) Miyamoto S, Akiyama Y, Nagata I, Karasawa J, Nozaki K, Hashimoto N, Kikuchi H: Long-term outcome after STA-MCA anastomosis for moyamoya disease. *Neurosurg Focus* 5: e5, 1998
9) Miyamoto S, Yoshimoto T, Hashimoto N, Okada Y, Tsuji I, Tominaga T, Nakagawara J, Takahashi JC; JAM Trial Investigators: Effects of extracranial-intracranial bypass for patients with hemorrhagic moyamoya disease: results of the Japan Adult Moyamoya Trial. *Stroke* 45: 1415–1421, 2014
10) Guzman R, Lee M, Achrol A, Bell-Stephens T, Kelly M, Do HM, Marks MP, Steinberg GK: Clinical outcome after 450 revascularization procedures for moyamoya disease. Clinical article. *J Neurosurg* 111: 927–935, 2009
11) 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* 100: 142–149, 2004

12) Kuroda S, Hashimoto N, Yoshimoto T, Iwasaki Y; Research Committee on Moyamoya Disease in Japan: Radiological findings, clinical course, and outcome in asymptomatic moyamoya disease: results of multicenter survey in Japan. *Stroke* 38: 1430–1435, 2007
13) Yamada M, Fujii K, Fukui M: [Clinical features and outcomes in patients with asymptomatic moyamoya disease—from the results of nation-wide questionnaire survey]. *No Shinkei Geka* 33: 337–342, 2005 (Japanese)
14) Nanba R, Kuroda S, Takeda M, Shichinohe H, Nakayama N, Ishikawa T, Houkin K, Iwasaki Y: [Clinical features and outcomes of 10 asymptomatic adult patients with moyamoya disease]. *No Shinkei Geka* 31: 1291–1295, 2003 (Japanese)
15) Baba T, Houkin K, Kuroda S: Novel epidemiological features of moyamoya disease. *J Neurol Neurosurg Psychiatr* 79: 900–904, 2008
16) Shinkawa A, Ueda K, Kiyohara Y, Kato I, Sueishi K, Tsuneyoshi M, Fujishima M: Silent cerebral infarction in a community-based autopsy series in Japan. The Hisayama Study. *Stroke* 26: 380–385, 1995
17) Ishikawa T, Kuroda S, Nakayama N, Terae S, Kudou K, Iwasaki Y: Prevalence of asymptomatic microbleeds in patients with moyamoya disease. *Neurol Med Chir (Tokyo)* 45: 495–500; discussion 500, 2005
18) Kikuta K, Takagi Y, Nozaki K, Hanakawa T, Okada T, Mikuni N, Miki Y, Fushmi Y, Yamamoto A, Yamada K, Fukuyama H, Hashimoto N: Asymptomatic microbleeds in moyamoya disease: T2*-weighted gradient-echo magnetic resonance imaging study. *J Neurosurg* 102: 470–475, 2005
19) Kikuta K, Takagi Y, Nozaki K, Okada T, Hashimoto N: Histological analysis of microbleed after surgical resection in a patient with moyamoya disease. *Neurol Med Chir (Tokyo)* 47: 564–567, 2007
20) Kikuta K, Takagi Y, Nozaki K, Sawamoto N, Fukuyama H, Hashimoto N: The presence of multiple microbleeds as a predictor of subsequent cerebral hemorrhage in patients with moyamoya disease. *Neurosurgery* 62: 104–111, discussion 111–112, 2008
21) Kuroda S, Kashiwazaki D, Ishikawa T, Nakayama N, Houkin K: Incidence, locations, and longitudinal course of silent microbleeds in moyamoya disease: a prospective T2*-weighted MRI study. *Stroke* 44: 516–518, 2013
22) Kuroda S, Ishikawa T, Houkin K, Nanba R, Hokari M, Iwasaki Y: Incidence and clinical features of disease progression in adult moyamoya disease. *Stroke* 36: 2148–2153, 2005
23) 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 Infractable Diseases: Guidelines for diagnosis and treatment of moyamoya disease (spontaneous occlusion of the circle of Willis). *Neurol Med Chir (Tokyo)* 52: 245–266, 2012

Appendix

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CASE REPORT

Headache attack followed by rapid disease progression in pediatric moyamoya disease—how should we manage it?

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Abstract
Case report A 4-year-old female was presented at our hospital with frequent right frontal headache attack. She was diagnosed with moyamoya disease and was conservatively followed up. One year later, the frequency of headache gradually decreased. However, follow-up MR imaging revealed that the disease stage markedly progressed in the right side and cerebral infarction occurred in the temporal lobe with atrophy of the right frontal lobe. She underwent direct and indirect revascularization on the right side.
Conclusion Aware of this case, we would like to emphasize that headache may be one subtype of ischemic attacks and require frequent MR follow-up to see the disease course. If there is any sign of disease progression, immediate surgical intervention should be indicated to avoid irreversible brain damage.

Keywords Moyamoya disease · Headache · Cerebral revascularization · Natural history · Disease progression

Introduction

Moyamoya disease is an uncommon cerebrovascular disease characterized by progress stenosis in the terminal internal carotid arteries (ICAs) and their main branches, which results in the formation of fine vascular networks at the base of the brain (moyamoya vessels) to compensate cerebral ischemia. Interestingly, moyamoya disease has two age distribution peaks at around 5 and 40 years. Most pediatric patients exhibit transient ischemic attack (TIA) or ischemic stroke. Headache and involuntary movements are also serious symptoms associated with pediatric moyamoya disease [1, 2]. Although the disease is rare, it is an important cause of ischemic stroke in children, and recently, it has become more widely recognized worldwide as a cause of pediatric cerebrovascular events. Thus, moyamoya disease should be considered in any child who presents with only headache attack.

In this report, we describe a pediatric case that presented with headache attacks due to moyamoya disease. During 1 year of follow-up, rapid disease progression led to cerebral infarction associated with intellectual impairment.

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Case

A 4-year-old female was presented at our hospital with frequent episodes of right frontal headache. She complained of it when she waked up in the morning and sometimes vomited. Physical and neurological examinations revealed no definite abnormality. Laboratory examinations, including erythrocyte sedimentation, somatic antibodies, and leptospiral antibodies, were all negative and ruled out any other underlying diseases. MR imaging (MRI) demonstrated no parenchymal lesion, but MR angiography (MRA) showed significant stenosis at the terminal portion of the right ICA and moyamoya vessels at the base of brain. There were no vascular lesions in the left side (Fig. 1). Therefore, we diagnosed as unilateral moyamoya disease. Except for the headache attacks, she was asymptomatic and her conditions seemed stable, so follow-up MR examination was scheduled 1 year later. One year later, she came back to the hospital for the follow-up. The frequency of headache attacks gradually decreased. However, MRA

revealed that the disease stage on the right side markedly progressed from stage 2 to stage 4 on Suzuki's grading. Right posterior cerebral artery was also involved. MRI demonstrated that cerebral infarction occurred in the right temporal lobe. There was also cerebral atrophy in the right frontal lobe (Fig. 1). 123I-IMP single photon emission computed tomography (SPECT) showed a marked reduction of cerebral blood flow in the right cerebral hemisphere. The intelligence quotient (IQ) was impaired. Therefore, she underwent superficial temporal artery to middle cerebral artery (STA-MCA) anastomosis combined with indirect bypass, encephalo-duro-myo-arterio-pericranial synangiosis (EDMAPS) [3, 4]. The frontal branch of STA was anastomosed to the prefrontal artery of MCA (0.8 mm in diameter). Temporary clamping time of MCA was 20 min. The patency of STA-MCA anastomosis was confirmed, using near infrared indocyanine green (ICG) videoangiography during surgery. The dura mater, temporal muscle, and frontal pericranium were used as the donor tissues for indirect bypass. Postoperative course was uneventful. She was discharged without any perioperative complications. During follow-up period, her headache attack almost disappeared. Repeated cerebral angiography revealed well-developed collaterals through both direct and indirect bypass (Fig. 2).

Discussion

Headache was not considered as an important clinical sign of pediatric moyamoya disease a decade ago. With the accumulation of clinical data, however, headache attack is now recognized as one of important initial symptoms [5-7]. Yamashiro et al. reported clinical feature of 10 Japanese children with moyamoya disease. The most common initial manifestations were headache in four cases (40 %) and motor deficit and convulsion in three cases (30 %) [7]. Battistella and Carollo described 34 Italian patients suffering from moyamoya disease. The early clinical symptoms consisted of TIA and/or stroke (44 %), recurrent migraine-like headache (20 %), seizure (18 %), and hemorrhage (3 %) [8]. Matsushima et al. reported that about ones-third of pediatric patients complained of headache at initial presentation. Headache significantly affected their activity of daily life (ADL) in more than 60 % of them. Majority of headache was localized in the frontal (40 %) or temporal region (25 %) [9]. Seol et al. also reported that headache was documented in 44 (21.6 %) of 204 pediatric patients with moyamoya disease. They also described that clinical course of headache in pediatric moyamoya disease had the following features: (1) a coexisting stage of headache and TIA; (2) the second stage of headache only; and (3) the final stage of improvement or disappearance of headache [6]. In fact, the frequency of headache gradually decreased during 1 year in the present case. Very recently, Kawabori et al. also reported the incidence and clinical features in pediatric moyamoya disease. Thus, headache attack was documented in 11 (38 %) of 29 pediatric patients. Severe disabling headache often develops in the frontal or temporal region in the morning but spontaneously resolves within 3 to 4 h [5].

Underlying mechanisms of headache in pediatric moyamoya disease is still unclear. Headache may arise from hypoperfusion-induced activation of pain-sensitive structures such as both intracranial and extracranial vasculatures, dura mater, and orbital and nasal cavities. Concomitantly, other mechanisms such as dilatation of the meningeal collaterals stimulating dural nocireceptors and ischemia-induced

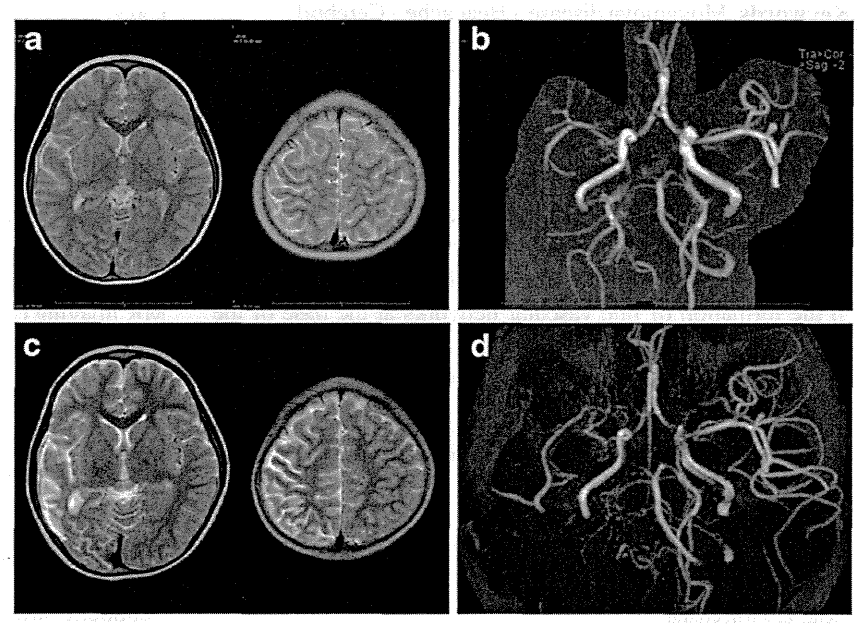

Fig. 1 T2-weighted MR imaging (**a**, **c**) and MR angiography (**b**, **d**). At initial presentation, there was no parenchymal lesion in the brain (**a**). The occlusive lesion in the right carotid termination was moderate (Suzuki's stage 2, **b**). One year later, however, cerebral infarction developed in the right temporal lobe. Cerebral atrophy was noted in the right frontal lobe (**c**). Disease stage markedly progressed in the right carotid termination to Suzuki's stage 4. The flow signal in the right posterior cerebral artery also decreased (**d**)

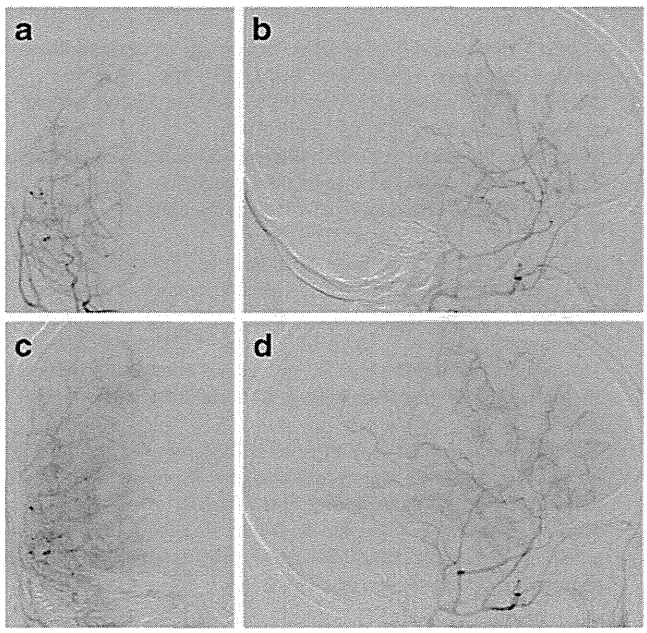

Fig. 2 Towne's view (**a**, **c**) and lateral view of postoperative right external carotid angiogram (**b**, **d**). Panels **a** and **b** represent early arterial phase, and panels **c** and **d** represent late arterial phase. They demonstrate that both direct and indirect bypass widely provide collateral blood flow to the right cerebral hemisphere

lowering of the migraine threshold have also been suggested [6]. Therefore, headache may develop as a symptom of TIA in pediatric moyamoya disease. Indeed, headache attack in pediatric patients is closely related to more advanced disease stage and also to decrease in cerebral blood flow and its reactivity to acetazolamide [5]. In the pediatric cases with atypical attacks of migraine and/or absence of a family history of migraine, therefore, a detailed investigation should be performed to detect underlying vascular diseases such as moyamoya disease.

Furthermore, there are a limited number of studies that denote the effect of surgical therapy on headache in pediatric moyamoya disease. Matsushima et al. reported that headache improved or disappeared in about 75 % of pediatric patients after encephalo-duro-arterio-synangiosis (EDAS) [9]. However, Seol et al. concluded that more than 60 % of pediatric patients still complained of headache even after EDAS [6]. Indeed, EDAS may provide no or little collateral blood flow to the frontal region where headache attacks often occur. Therefore, the impact of EDAS on headache attack is still controversial in pediatric moyamoya disease. On the other hand, very recent report has shown that STA-MCA anastomosis combined with EDMAPS markedly improves cerebral hemodynamics and resolves headache attacks in all of 11 pediatric patients with moyamoya disease, suggesting the importance of surgical design through a large craniotomy covering the frontal area [5]. It is still obscure whether surgical revascularization would be beneficial for pediatric patients with only headache attack due to moyamoya disease. However, there is the possibility that surgical revascularization at initial presentation may possibly prevent cerebral infarction in the present case. Further studies are warranted to clarify this issue. Well-designed surgical treatment should be indicated without delay, at least when the pediatric patients with only headache due to moyamoya disease shows disease progression or deterioration of cerebral hemodynamics on follow-up examinations. Future clinical testing would be warranted to resolve this issue.

In the present case, disease stage markedly progressed during 1 year of follow-up, causing irreversible brain damage. Occlusive lesions in the carotid terminations often worsen in pediatric moyamoya disease [10, 11]. In fact, Fukuyama and Umezu reported that occlusive lesions progressed in about two-thirds of hemispheres on repeat cerebral angiography with a mean interval of about 3 years in pediatric moyamoya disease [12]. Therefore, the radiological course in the present case may be natural, although the incidence of disease progression during 1 year is still unknown. Based on these observations, follow-up MR examination should be planned with shorter interval, for example, every 3 to 6 months. In addition, it is well known that functional and/or intellectual outcomes are poor in pediatric patients presenting with ischemic stroke due to moyamoya disease [13-15]. To prevent the occurrence of cerebral infarction, therefore, regular imaging studies should also be planned with shorter intervals in the conservatively treated children with moyamoya disease, because MRI and MRA studies enable it to accurately detect disease progression at outpatient clinic [16]. Surgical revascularization should also be indicated in the cases that show rapid disease progression in order to prevent permanent neurological and cognitive deficits.

In summary, we describe a case of pediatric moyamoya disease that presented with headache attacks at initial presentation. During 1 year of follow-up, a rapid disease progression caused cerebral infarction associated with cognitive impairment. We are convinced that a closer radiological monitoring would identify the disease progression more promptly and prevent permanent neurological sequelae due to ischemic stroke by indicating surgical revascularization.

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References

1. Kuroda S, Houkin K (2008) Moyamoya disease: current concepts and future perspectives. Lancet Neurol 7:1056–1066
2. Suzuki J, Takaku A (1969) Cerebrovascular "moyamoya" disease. Disease showing abnormal net-like vessels in base of brain. Arch Neurol 20:288–299

3. 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
4. Kuroda S, Houkin K (2012) Bypass surgery for moyamoya disease: concept and essence of surgical technique. Neurol Med Chir (Tokyo) 52:287–294
5. Kawabori M, Kuroda S, Nakayama N, Hirata K, Shiga T, Houkin K, Tamaki N (2013) Effective surgical revascularization improves cerebral hemodynamics and resolves headache in pediatric moysamoya disease. World Neurosurg 80:612–619
6. Seol HJ, Wang KC, Kim SK, Hwang YS, Kim KJ, Cho BK (2005) Headache in pediatric moyamoya disease: review of 204 consecutive cases. J Neurosurg 103:439–442
7. Yamashiro Y, Takahashi H, Takahashi K (1984) Cerebrovascular moyamoya disease. Eur J Pediatr 142:44–50
8. Battistella PA, Carollo C (1997) Clinical and neuroradiological findings of moyamoya disease in Italy. Clin Neurol Neurosurg 99(Suppl 2):S54–S57
9. Matsushima Y, Aoyagi M, Nariai T, Nojiri T, Ohno K (2000) Headache in pediatric moyamoya patients: pre- and postoperative changes. Nerv Syst Child (Jpn) 25:442–447
10. Ezura M, Yoshimoto T, Fujiwara S, Takahashi A, Shirane R, Mizoi K (1995) Clinical and angiographic follow-up of childhood-onset moyamoya disease. Childs Nerv Syst 11:591–594
11. Ibrahimi DM, Tamargo RJ, Ahn ES (2010) Moyamoya disease in children. Childs Nerv Syst 26:1297–1308
12. Fukuyama Y, Umezu R (1985) Clinical and cerebral angiographic evolutions of idiopathic progressive occlusive disease of the circle of Willis ("moyamoya" disease) in children. Brain Dev 7:21–37
13. 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
14. Kurokawa T, Tomita S, Ueda K, Narazaki O, Hanai T, Hasuo K, Matsushima T, Kitamura K (1985) Prognosis of occlusive disease of the circle of Willis (moyamoya disease) in children. Pediatr Neurol 1: 274–277
15. Veeravagu A, Guzman R, Patil CG, Hou LC, Lee M, Steinberg GK (2008) Moyamoya disease in pediatric patients: outcomes of neurosurgical interventions. Neurosurg Focus 24:E16
16. Kuroda S, Ishikawa T, Houkin K, Nanba R, Hokari M, Iwasaki Y (2005) Incidence and clinical features of disease progression in adult moyamoya disease. Stroke 36:2148–2153

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Case Report

Early onset of moyamoya syndrome in a Down syndrome patient with the genetic variant *RNF213* p.R4810K

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Abstract

Moyamoya syndrome is a unique progressive occlusive cerebrovascular disease that predisposes affected patients to stroke. We describe the case of a 2-year-old girl presenting with early onset of moyamoya syndrome with concurrent Down syndrome. Genetic testing revealed a heterozygous missense variant of *RNF213*. *RNF213* was recently identified as the first susceptibility gene for moyamoya disease in patients with no known associated risk factors. The reported median age at the onset of idiopathic moyamoya disease with a heterozygous *RNF213* risk variant is 7 years, while, the average age at onset of moyamoya syndrome in Down syndrome is 7–16 years. Down syndrome and *RNF213* variant contribute to the development of moyamoya vasculopathy in different ways. Although the underlying mechanism is not fully understood, an additive effect was observed with the early-onset seen in this patient. Little is known about the potential association between *RNF213* and moyamoya syndrome. Based on these observations, we hypothesize that the *RNF213* risk variant has a modifier effect in steno-occlusive vasculopathy, even in medical conditions known to be associated with moyamoya syndrome.

Keywords: Moyamoya syndrome; Down syndrome; *RNF213*; Pediatric stroke

1. Introduction

Moyamoya syndrome (MMS) is a chronic cerebrovascular condition that predisposes affected patients to stroke in association with progressive stenosis of the intracranial internal carotid arteries and their proximal branches. Proliferation of fine vascular collaterals around the occlusive region is a distinctive feature of this syndrome, and gives rise to a "puff of smoke" radiographic appearance, which is termed "moyamoya" in Japanese. Patients who have the characteristic moyamoya vasculopathy and also have other associated conditions (for example, Down syndrome (DS) and neurofibromatosis type 1) are categorized as having MMS. Whereas, patients with no known associated risk factors are described as having moyamoya disease (MMD) [1].

The etiology of MMD is probably multifactorial, but several lines of evidence support the importance of genetic factors in susceptibility to MMD. Twelve percent of individuals with MMD have a family history of the disease [2]. The *RNF213* gene on the 17q25.3 locus was recently identified as the first susceptibility gene for MMD [3,4], and the p.R4810K missense variant (rs 112735431, G>A) has been reported to increase the risk

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of MMD with an odds ratio of 190.8–338.9 in the Japanese population [3–5]. The median age of onset in patients with the heterozygous *RNF213* pattern was found to be 7 years [5]. Herein, we report the case of a 2-year-old Japanese patient who had early-onset MMS with concurrent DS and had the heterozygous *RNF213* missense variant.

2. Clinical report

A Japanese girl presenting with muscle hypotonia, epicanthal folds, flattened nose, and congenital heart disease at birth was diagnosed with DS. Her chromosome analysis using G banding karyotyping showed 47,XX,+21. The patient underwent atrial and ventricular septal defect patch closure at the age of 1 month, and remained symptom free after the procedure.

At the age of 2 years and 9 months, the patient was referred to us because of weakness in her left arm. Neurologic examination revealed left facial hemiparesis and 4/5 motor strength with manual muscle testing in the left upper extremity. Magnetic resonance imaging of the brain showed a fresh cerebral infarction in the right frontal cortex as well as old infarction lesions in the deep white matter (Fig. 1A and B). An intracranial magnetic resonance angiogram revealed the characteristic steno-occlusive features and accompanying collaterals of MMS (Fig. 2). Evaluation of coexisting autoimmune disorders including immunological and array of autoantibodies studies were negative.

The patient was treated conservatively. Oral aspirin therapy was commenced after discharge. She was later referred to the neurosurgery department for surgical intervention evaluation, and a bypass procedure was scheduled for a later date. The patient received genetic testing for *RNF213*. Genotyping was conducted using

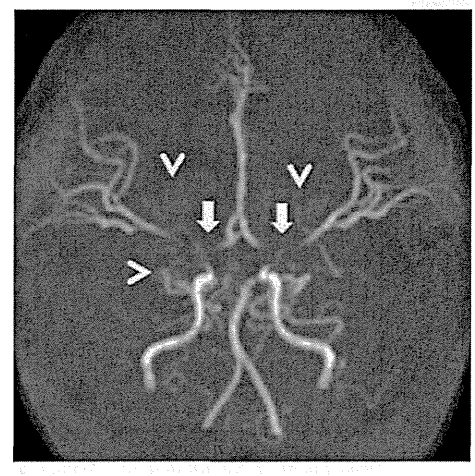

Fig. 2. Intracranial magnetic resonance angiography revealing bilateral stenosis of the terminal portion of the internal carotid artery, bilateral occlusion of the proximal region of the middle cerebral artery (*arrow*), and a fine network of collaterals (*arrowhead*) in proximity.

a TaqMan® SNP Genotyping assay (Applied Biosystems, Foster City, CA) and revealed a heterozygous pattern in the p.R4810K variant [3]. Genotyping of the patient's father also showed the same heterozygous pattern. The father is asymptomatic and has no headache, paralysis or other complaints. 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.

3. Discussion

The p.R4810K variant has been identified as a founder variant commonly observed in Japanese and other East Asian patients. Mutational analyses of the *RNF213* gene have revealed a p.R4810K variant in

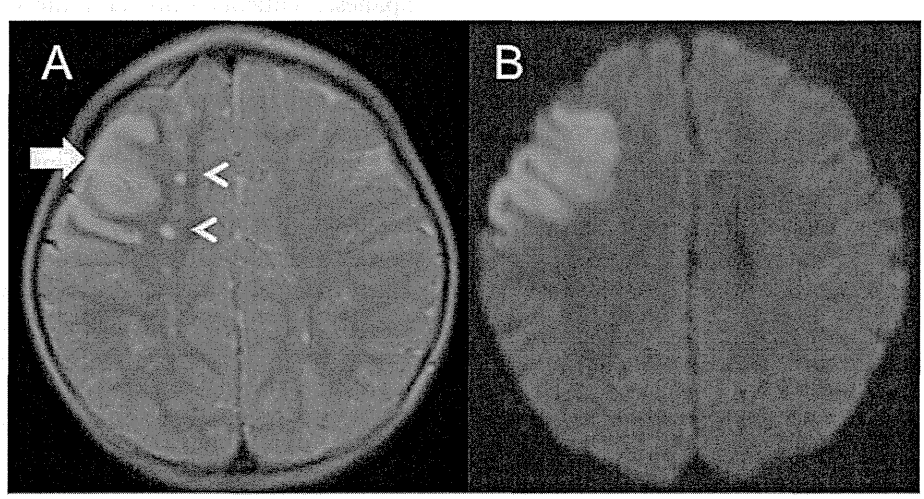

Fig. 1. Brain magnetic resonance imaging during admission. (A) Axial T2-weighted image showing high intensity area in the right frontal cortex (*arrow*) and old infarction lesions (*arrowhead*) in the deep white matter. (B) MRI diffusion-weighted imaging sequence confirming a fresh cerebral infarction of the right frontal cortex.

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95.0–95.1% of familial MMD cases, 73.0–79.2% of non-familial MMD cases, and 1.4–1.8% of controls [4,5]. A gene dosage effect was observed with the homozygous variant showing an earlier onset age and a more severe form of the disease in comparison to the heterozygous variant [5]. The median age at onset in Japanese MMD patients was found to be 3, 7, and 8 years for homozygotes, heterozygotes, and wild types, respectively [5]. The MMS onset age in the DS patient showing a heterozygous pattern of the *RNF213* variant as described in this study was 2 years, much earlier than the reported onset age of MMD with a heterozygous variant. Although the clinical effect of *RNF213* in MMS that occurs together with recognizable medical conditions is unknown, our observations lead us to hypothesize that the *RNF213* risk variant may further promote and accelerate the vasculopathy in a subset of populations highly susceptible to MMS.

A higher prevalence of MMS in DS patient than in general populations is commonly observed. Based on data from the Nationwide Inpatient Sample, the estimated prevalence of coexisting MMD and DS is 3.8% (3760 per 100,000) among patients admitted with MMD in the United States [6]. The prevalence of DS in patients with coexisting MMS was 26-fold greater than the prevalence of DS among live births (145 per 100,000) [6]. In Japan, the estimated prevalence of MMS in DS patients is estimated to be one in 400–532 [7], whereas the prevalence of MMD in the general population is estimated to be one in 15,000 [2,7]. In every racial or ethnic group, therefore, the occurrence of MMS in patients with DS may be higher by a similar proportion compared to idiopathic MMD in general populations. In a series of case reports, the average age at onset of MMS was reported to be 7 years [8]; the mean admission age was found to be 16 years in a large observational study [6]. The present 2 year-old DS patient, by having a *RNF213* variant, observes an accelerated onset age of MMS. This may suggest that DS and *RNF213* variant contribute to the development of moyamoya vasculopathy, with different mechanisms showing an additive effect.

Recent studies have revealed that the *RNF213* gene may play a role in angiogenesis [9], vascular development [3], and mitosis [10]. In experimental medicine, overexpression of *RNF213* p.R4810K in HeLa cells extended the time of mitosis by 4-fold [10]. Aneuploidy was observed more frequently in induced pluripotent stem cells and fibroblasts from patients with MMD carrying *RNF213* mutation [10]. Thus, *RNF213* variant may induce mitotic abnormalities and chromosomal instability. The molecular mechanism of vascular occlusion in DS with MMS is unknown; however, it has been postulated that several proteins encoded on chromosome 21 (α-chains of collagen type VI, interferon-γ receptor and others) may be inappropriately expressed, causing an increased risk of moyamoya vasculopathy [6,8]. Further study on the cross-talk between *RNF213* and conditions that cause MMS is warranted to foster a better understanding of these molecular mechanisms.

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References

[1] Scott RM, Smith ER. Moyamoya disease and moyamoya syndrome. N Engl J Med 2009;360:1226–37.
[2] Kuriyama S, Kusaka Y, Fujimura M, Wakai K, Tamakoshi A, Hashimoto S, et al. Prevalence and clinicoepidemiological features of moyamoya disease in Japan: findings from a nationwide epidemiological survey. Stroke 2008;39:42–7.
[3] 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.
[4] 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 2011;56:34–40.
[5] Miyatake S, Miyake N, Touho H, Nishimura-Tadaki A, Kondo Y, Okada I, et al. Homozygous c.14576G>A variant of RNF213 predicts early-onset and severe form of moyamoya disease. Neurology 2012;78:803–10.
[6] Kainth DS, Chaudhry SA, Kainth HS, Suri FK, Qureshi AI. Prevalence and characteristics of concurrent down syndrome in patients with moyamoya disease. Neurosurgery 2013;72:210–5.
[7] Fukushima Y, Kondo Y, Kuroki Y, Miyake S, Iwamoto H, Sekido K, et al. Are Down syndrome patients predisposed to moyamoya disease? Eur J Pediatr 1986;144:516–7.
[8] Dai AI, Shaikh ZA, Cohen ME. Early-onset moyamoya syndrome in a patient with Down syndrome: case report and review of the literature. J Child Neurol 2000;15:696–9.
[9] Hitomi T, Habu T, Kobayashi H, Okuda H, Harada KH, Osafune K, et al. Downregulation of Securin by the variant RNF213 R4810K (rs112735431, G>A) reduces angiogenic activity of induced pluripotent stem cell-derived vascular endothelial cells from moyamoya patients. Biochem Biophys Res Commun 2013;438:13–9.
[10] Hitomi T, Habu T, Kobayashi H, Okuda H, Harada KH, Osafune K, et al. The moyamoya disease susceptibility variant RNF213 R4810K (rs112735431) induces genomic instability by mitotic abnormality. Biochem Biophys Res Commun 2013;439:419–26.

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特集

Ⅱ. 脳卒中と遺伝子 Update

もやもや病と *RNF213* 遺伝子

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われわれは *RNF213* 遺伝子がもやもや病の感受性遺伝子であること，p.R4810K 多型が感受性多型であることを報告した．近年の機能解析により，*RNF213* R4810K は Securin の発現低下を通じて血管内皮細胞の血管形成能を低下させること，*MAD2* 機能阻害を通じて細胞分裂異常およびゲノム不安定性を引き起こすことが明らかになり，これらの機能異常がもやもや病の発症機構に関与すると考えられる．

Key Words
もやもや病，*RNF213*，iPS 細胞，血管内皮細胞，細胞分裂異常

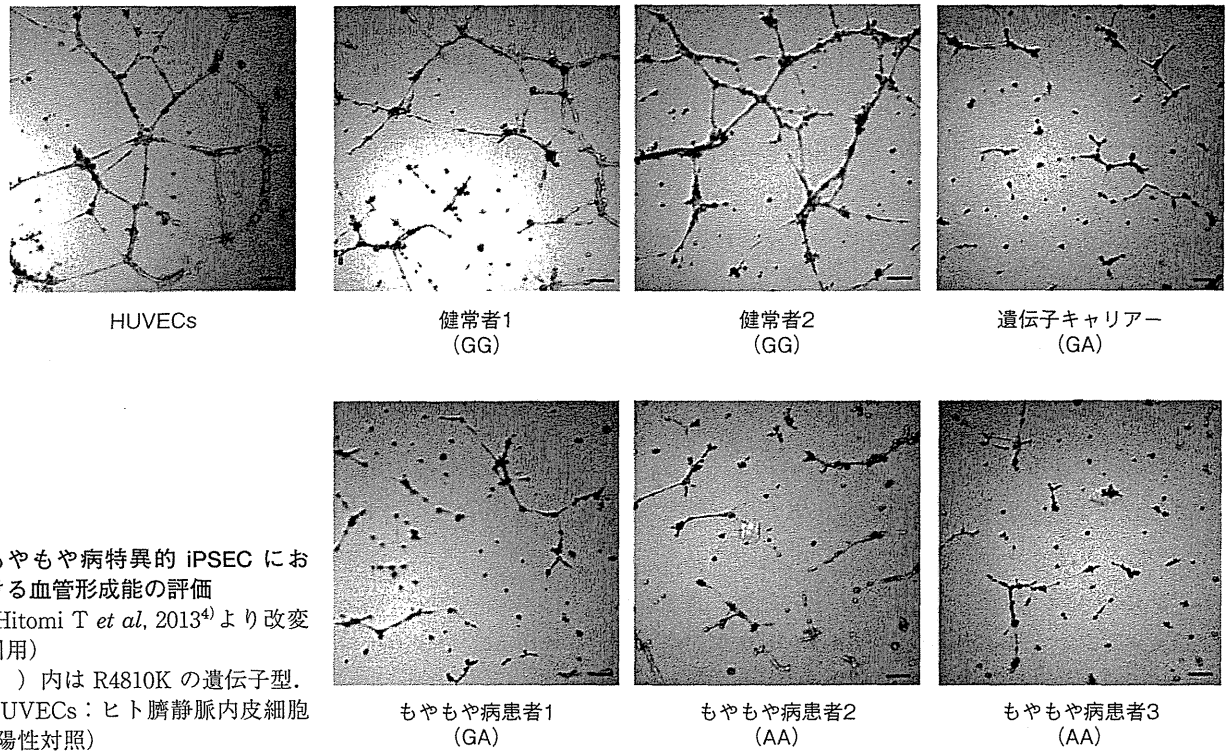
はじめに

もやもや病は内頸動脈終末部の狭窄および代償的な異常血管網（もやもや血管）の形成を特徴とする脳血管疾患である．もやもや病はわが国をはじめ東アジア地域で多発することが以前より知られており，さらに近年では欧米での報告も相次ぎ，世界中で小児脳血管障害の主たる原因として認められている．本稿では近年われわれが明らかにしたもやもや病の感受性遺伝子 *RNF213* の遺伝解析および機能解析について概説する．

1　もやもや病感受性遺伝子 *RNF213* の同定

もやもや病は東アジアで多発する点，患者の約 15%に家族歴を認める点から発症に遺伝素因が関与することが想定されてきたが，原因遺伝子は不明であった．われわれはもやもや病に関与する遺伝因子を明らかにするために，常染色体優性遺伝家系を収集し，全ゲノム連鎖解析およびエクソーム解析をおこなった．その結果，17q25.3 に存在する *RNF213* 遺伝子の 4,810 番目のアルギニンがリジンへと変化する多型（p.R4810K，現在は dbSNP に rs112735431 で登録*）が家系内のもやもや病患者に共通して見出されることを示した[1]．日中韓の孤発性もやも

*現在 NCBI に登録されている p.R4810K はわれわれがおこなったクローニング結果のみならず，他種動物との相同性の観点からも採用されている．p.R4859K の表記も散見されるが，Bioinformatics により予想される産物に基づいており実験的根拠に乏しい．



図❶ もやもや病特異的 iPSEC における血管形成能の評価
(Hitomi T *et al*, 2013[4]より改変引用)
( ) 内は R4810K の遺伝子型.
HUVECs：ヒト臍静脈内皮細胞(陽性対照)

や病患者における相関解析でも p.R4810K は患者で高頻度に認め（日本：90%，韓国：79%，中国：23%），3 ヵ国全体では p.R4810K による発症リスクは約 112 倍となった[1]．また一方で，東アジアの一般人口のうち約 2〜3%に p.R4810K を有する未発症の遺伝子キャリアーが存在することが示され，東アジア全体ではハイリスクな遺伝子キャリアーは 1,500 万人を超えると推計される．さらに *RNF213* のクローニングをおこなった結果，米国生物工学情報センター（NCBI）登録情報と遺伝子構造に差異があり，さらに実験的に産生した 591 kDa 蛋白の *in vitro* での機能解析により Walker A および B モチーフと RING finger 領域をもつ蛋白であることが証明されたため，新規蛋白として mysterin（moyamoya stenoocclusive disease-associated AAA+and RING finger protein）と命名した．また，近年の Morito ら[2]の研究では新規の AAA+ATPase であることが示されている．さらにゼブラフィッシュ，マウスの *RNF213* 欠損モデルを用いた実験では血管発生および小胞体ストレス応答に重要な役割を果たすことが明らかとなった[1)3)]．しかし，*RNF213* 欠損マウスでは，血管病変は認められず[3]，もやもや病の病態における役割は未解明な部分が多い．

2 もやもや病特異的 iPS 細胞由来血管内皮細胞の血管形成能の低下

もやもや病における *RNF213* R4810K の役割を明らかにする目的で，R4810K をヘテロ(GA)あるいはホモ(AA)で有するもやもや病患者および遺伝子キャリアーより iPS 細胞を作製し，血管内皮細胞（iPS endothelial cell：iPSEC）への分化をおこなった．Tube formation assay による血管形成能の評価をおこなった結果，もやもや病患者および遺伝子キャリアー由来の iPSEC では血管形成能が低下していることが明らかとなった（図❶）[4]．またマイクロアレイ解析の結果，患者および遺伝子キャリアー由来の iPSEC では発現プロファイルに健常者と比較して顕著な違いがあり，とくに細胞分裂に関与する多数の遺伝子群が低下していることが示された．

見出された iPSEC の表現型が *RNF213* R4810K の過剰発現により再現されるか検証するために，ヒト臍静脈内皮細胞（human unbilical vein endothelial cell：HUVEC）を用いて *RNF213* R4810K の強制発現をおこなった．その結果，*RNF213* R4810K 強制発現細胞は血管形成能の低下を示したが，それに対して siRNA による *RNF213* 抑制は血管形成能に影響を与えなかった．この結果より

RNF213 R4810K は gain of function の形式で細胞機能に影響を与えることが示唆された．われわれは血管形成能の低下への Securin の関与に着目した．Securin は患者由来 iPSEC で低下していた細胞分裂関連遺伝子群のうちの一つで，未成熟な姉妹染色体の分離を防ぎ，また血管新生においては細胞の遊走機能を介して血管形成に重要な役割を果たすことが知られている．Securin は患者由来 iPSEC と同様に *RNF213* R4810K 強制発現細胞でも低下していることが明らかになり，さらに siRNA による Securin 抑制は HUVEC および iPSEC において血管形成を低下させることが示された．

以上より，*RNF213* R4810K は Securin の発現低下を通じて血管内皮細胞の血管形成能を低下させることで，もやもや病の病態に関与することが示唆された．またもやもや病患者からの iPS 細胞樹立は現在まで報告がなく，もやもや病 *in vitro* モデルとしての利用も期待できる．

3 *RNF213* R4810K 強制発現による細胞分裂機能への影響

もやもや病患者由来 iPSEC において細胞分裂関連遺伝子の発現低下を認めた点から，*RNF213* R4810K を強制発現したヒト子宮頸癌細胞 HeLa 細胞を用いて細胞分裂機能の評価をおこなった．*RNF213* R4810K の過剰発現により，細胞増殖が大きく低下し，また細胞周期 M 期は約 4 倍に延長し，mitotic failure の頻度が上昇していることが明らかになった[5]．

さらに *RNF213* と M 期制御に主要な役割を果たす Mitotic arrest deficiency2 (*MAD2*) についての解析をおこなった．M 期前中期において，*MAD2* は正常では染色体の動原体に存在する．それに対して，*RNF213* R4810K 発現細胞では *MAD2* は *RNF213* R4810K と共局在を示し，動原体には存在しなかった．また，免疫沈降法により *RNF213* は *MAD2* と複合体形成を認め，*RNF213* R4810K は野生型と比較してより強く複合体を形成することが示された．この結果は *RNF213* R4810K が *MAD2* への吸着により正常な局在を阻害することを示唆している．さらに p.R4810K を有するもやもや病患者から線維芽細胞を単離し M 期停止を誘導するノコダゾールで処理して解析をおこなった結果，患者由来線維芽細胞においては，健常者由来線維芽細胞と比較し，*MAD2* の異常な局在が観察された．さらに染色体異数性が有意に増加していることが明らかになり，R4810K がゲノム不安定性を導くことが示唆された．

おわりに

本稿では，もやもや病の感受性遺伝子である *RNF213* 遺伝子の p.R4810K 多型が Securin の発現低下を通じて血管内皮細胞の血管形成能を低下させること，MAD2 機能阻害を通じて細胞分裂異常およびゲノム不安定性を引き起こすことを述べた．*RNF213* R4810K はもやもや病において，①血管内皮細胞の機能異常，②細胞分裂異常・ゲノム不安定性を通じた血管内皮細胞死を引き起こし，脳血管の狭窄につながると考えられる．

●文　献●

1) Liu W *et al*：Identification of RNF213 as a susceptibility gene for moyamoya disease and its possible role in vascular development. *PLoS One* **6**：e22542, 2011
2) Morito D *et al*：Moyamoya disease-associated protein mysterin/RNF213 is a novel AAA＋ATPase, which dynamically changes its oligomeric state. *Sci Rep* **4**：4442, 2014
3) Kobayashi H *et al*：Ablation of Rnf213 retards progression of diabetes in the Akita mouse. *Biochem Biophys Res Commun* **432**：519-525, 2013
4) Hitomi T *et al*：Downregulation of Securin by the variant RNF213 R4810K (rs112735431, G>A) reduces angiogenic activity of induced pluripotent stem cell-derived vascular endothelial cells from moyamoya patients. *Biochem Biophys Res Commun* **438**：13-19, 2013
5) Hitomi T *et al*：The moyamoya disease susceptibility variant RNF213 R4810K (rs112735431) induces genomic instability by mitotic abnormality. *Biochem Biophys Res Commun* **439**：419-426, 2013

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Genetic Study of Intracranial Aneurysms

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Background and Purpose—Rupture of intracranial aneurysms (IAs) causes subarachnoid hemorrhage, leading to immediate death or severe disability. Identification of the genetic factors involved is critical for disease prevention and treatment. We aimed to identify the susceptibility genes for IAs.

Methods—Exome sequencing was performed in 12 families with histories of multiple cases of IA (number of cases per family ≥3), with a total of 42 cases. Various filtering strategies were used to select the candidate variants. Replicate association studies of several candidate variants were performed in probands of 24 additional IA families and 426 sporadic IA cases. Functional analysis for the mutations was conducted.

Results—After sequencing and filtering, 78 variants were selected for the following reasons: allele frequencies of variants in 42 patients was significantly ($P<0.05$) larger than expected; variants were completely shared by all patients with IA within ≥1 family; variants predicted damage to the structure or function of the protein by PolyPhen-2 (Polymorphism Phenotyping V2) and SIFT (Sorting Intolerance From Tolerant). We selected 10 variants from 9 genes (*GPR63*, *ADAMST15*, *MLL2*, *IL10RA*, *PAFAH2*, *THBD*, *IL11RA*, *FILIP1L*, and *ZNF222*) to form 78 candidate variants by considering commonness in families, known disease genes, or ontology association with angiogenesis. Replicate association studies revealed that only p.E133Q in *ADAMTS15* was aggregated in the familial IA cases (odds ratio, 5.96; 95% confidence interval, 2.40–14.82; $P=0.0001$; significant after the Bonferroni correction [$P=0.05/78=0.0006$]). Silencing *ADAMTS15* and overexpression of *ADAMTS15* p.E133Q accelerated endothelial cell migration, suggesting that *ADAMTS15* may have antiangiogenic activity.

Conclusions—*ADAMTS15* is a candidate gene for IAs. **(*Stroke*. 2015;46:620-626. DOI: 10.1161/STROKEAHA.114.007286.)**

Key Words: genetics ■ intracranial aneurysms

The overall prevalence of intracranial aneurysms (IAs) is estimated at 3.2% in the general population.[1] The rupture of an IA is one of the most devastating neurological conditions known.[2] Multiple risk factors, such as cigarette smoking, hypertension, and alcohol consumption, are known risk factors for the formation and possible rupture of IAs.[3–5] The familial occurrence of IAs suggests that genetic factors are involved in disease susceptibility.[6,7] Although several genome-wide association studies have been performed worldwide[8–13] because of their limited power to detect rare variants that are thought to have larger effect sizes, the genetic predisposition of IAs is largely unknown.

In accordance with the hypothesis that less common variants (minor allele frequency [MAF] <0.05) may contribute to IA development, we performed whole exome sequencing of 42 cases with a definite phenotype of IA from 12 families. Further replicate association studies of several candidate variants in additional familial and sporadic IA cases and biological investigation were performed.

Methods

Study Population

In cooperation with hospitals in the Western part of Japan, we established a large cohort of familial and sporadic IA cases.[14] These cases were diagnosed by angiography or subarachnoid hemorrhage with aneurysm rupture or in some cases, unruptured IAs were confirmed during intracranial surgery. Individual information and lifestyle data were collected by interview. With respect to familial IA, these families showed high IA aggregation (≥2 first- to third-degree relatives affected by IA) with a supposed strong genetic component. Twelve families with ≥3 definite IA cases (a total of 42 cases) were selected for whole exome sequencing (Figure 1). The probands of another unrelated 24 IA families were used for further replication study. Then, 426 unrelated sporadic IA cases were used for further association study of the selected candidate variants. Written informed consent was obtained from all participants.

For the control population, we used exome data from the Japanese genetic variation consortium database (a reference database of genetic variations in the Japanese population that contains genetic variations determined by exome sequencing of 1208

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