

A novel CCM3 mutation associated with cerebral cavernous malformation in a Chinese family

Xiao-yu Jiang , Ying Zhang, Xiang Yin, Di Nan, Xu Wang, Jia-chun Feng and Jing Miao

Ther Adv Neurol Disord

2020, Vol. 13: 1–6

DOI: 10.1177/
1756286420902664

© The Author(s), 2020.
Article reuse guidelines:
sagepub.com/journals-
permissions

Abstract

Background: Cerebral cavernous malformation (CCM), especially the familial form, is a relatively rare congenital and occult vascular disease of the central nervous system. The familial form of CCM has been linked to three different genes: *KRIT1/CCM1*, *MGC4607/CCM2*, and *PDCD10/CCM3*; however, the genetic basis of CCM is not well understood. The *PDCD10/CCM3* is the most recent gene to be identified that results in worse clinical symptoms. Early diagnosis and treatment is important for patient prognosis.

Case report: The proband is a 38-year-old male who has been suffering from weakness in the limbs for 7 months. Investigation of his family history revealed that his mother also suffered from limbs paralysis and had been bedridden for a long time. His older brother suffered from headache for years, whereas his younger brother was asymptomatic. Brain computed tomography analysis of all family members showed multiple high-density shadows. Subsequently, magnetic resonance imaging analysis identified more prominent and similar multiple intracranial lesions in all family members. The lesions were hypo-intense, or showed mixed signs on T1-weighted imaging, and were significantly more intense on T2-weighted imaging. To understand the genetic basis of the disease in the family, DNA sequencing analysis was performed. A novel deletion mutation in the *PDCD10/CCM3* gene was identified in the proband and his relatives. The deletion resulted in a frameshift mutation and premature termination of translation of the protein, and potentially caused the disease in this family.

Conclusions: Our study identified a novel *PDCD10/CCM3* heterozygous deletion (c.165delT) associated with CCM. This finding expands the *CCM* gene mutation profile, which will be beneficial for genetic counseling and clinical therapy.

Keywords: autosomal dominant inheritance, cerebral cavernous malformation, gene mutation, *PDCD10/CCM3*, vascular disease

Received: 21 October 2019; revised manuscript accepted: 17 December 2019.

Introduction

Cerebral cavernous malformation (CCM) is classically defined as low-pressure, hamartomatous, and berry-like vascular lesions in the central nervous system. The prevalence of all types of CCM is predicted to be 0.1–0.5% in the general population.^{1,2} CCM occurs either as sporadic or familial; inheritance of the familial form is autosomal dominant, with variable expression and incomplete penetrance.³ It is clinically silent, or associated with

headache, hemorrhagic strokes, seizures, or focal neurological deficits. In addition to the symptoms, brain computed tomography (CT) and magnetic resonance imaging (MRI) are commonly used for examining CCM, with brain MRI being more sensitive in revealing lesions, especially on susceptibility weighted imaging (SWI). Images of CCM do not appear the same in all individuals due to the repeated episodes of bleeding in brain lesions. A typical image can be seen on T2-weighted MRI of

Correspondence to:
Jing Miao
Jilin University First
Hospital, Changchun, Jilin,
China, 130021

475764792@qq.com

Jia-chun Feng
The First Hospital of
Jilin University, China
fengjcfanki@qq.com

Xiao-yu Jiang
Ying Zhang
Xiang Yin
Di Nan
Xu Wang
First Hospital, Jilin
University, Changchun,
Jilin, China

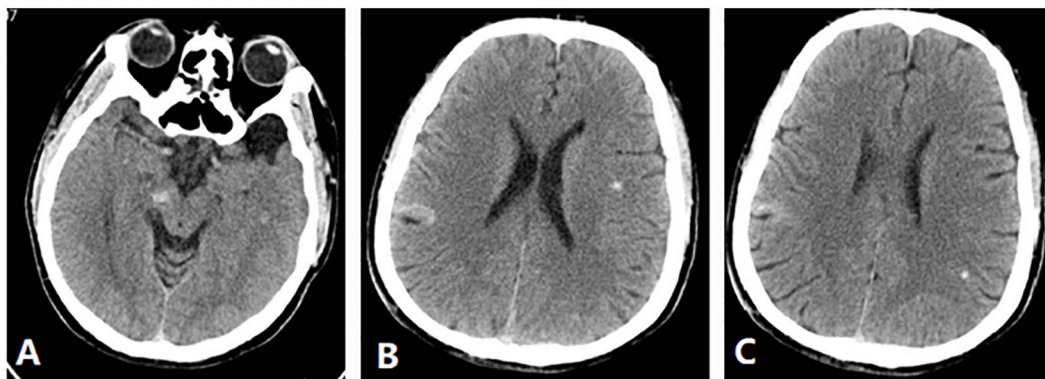


Figure 1. Brain CT of the proband shows small high-density shadows in the cerebral peduncle (a), cortical/juxta-cortical region and corona radiata (b, c). CT, computed tomography.

the brain, which is characterized by a mulberry- or popcorn-like appearance in the center, surrounded by a dark rim.⁴ CCM is caused genetically by loss-of-function mutations in one of the following three genes: *KRIT1* (Krev1 Interaction Trapped 1)/*CCM1*, *MGC4607* (malcavermin)/*CCM2*, and *PDCD10* (Programmed Cell Death 10)/*CCM3*. To date, more than 100, 30, and 20 distinct mutations have been identified in *CCM1*, *CCM2*, and *CCM3*, respectively.¹ In our study, a novel mutation in the *CCM3* gene was identified in a Chinese family. This finding widens our understanding of the mutation pools in genes associated with CCM, which will be useful for studying CCM in the future.

Case report

A 38-year-old male presented to our outpatient department with a 7-month history of left limbs weakness. He had no history of smoking, hypertension, diabetes, or any other risk factors for cerebrovascular diseases. Physical examination upon admission revealed paralysis of the left limbs and slight paralysis of the face. Babinski and Chaddock signs on the left side were positive, whereas other nervous system examination results were negative. Brain CT showed multiple high-density shadows, which indicated a tendency for calcification (Figure 1). For further diagnosis, MRI was performed, and more lesions were found with MRI compared with CT. The brain lesions were partly hypo-intense on T1-weighted imaging, whereas they were significantly more intense on T2-weighted imaging. The other lesions were hyper-intense in the center and surrounded by a

hypo-intense rim on T1/T2-weighted imaging. SWI displayed more hypo-intense lesions, indicating hemorrhage (Figure 2). Investigation of the family history showed that his relatives also suffered from physical disorders, albeit to different degrees. His mother suffered from progressive limb paralysis, and had been bedridden for a long time. Her brain MRI showed leukoaraiosis and multiple hypo-intense lesions on T2/T1 and diffusion weighted imaging (DWI), indicating hemorrhage. His older brother had suffered from headache for several years without any symptoms of limb paralysis. His younger brother was asymptomatic. Although his brothers had different symptoms, we noted similar findings on the brain CT and MRI (Figure 3) scans. All observations indicated that the patient probably had a genetic disease.

Molecular genetics studies

To examine the genetic cause of the disease, after obtaining written informed consent from the family members, DNA sequencing was performed. Genomic DNAs were extracted from the peripheral venous blood of all the family members using a blood DNA extraction Kit (CWE9600). DNA was prepared in the laboratory using a KAPA Library Preparation Kit; sequencing was then performed using Illumina NovaSeq. Sequence analysis of the *CCM1*, *CCM2*, and *CCM3* genes identified a novel heterozygous deletion in the *CCM3* gene (c.165delT, p.N55fs) (Figure 4). The 165th nucleotide in the coding region of the *CCM3* gene had a heterozygous deletion, which resulted in a frameshift mutation leading to

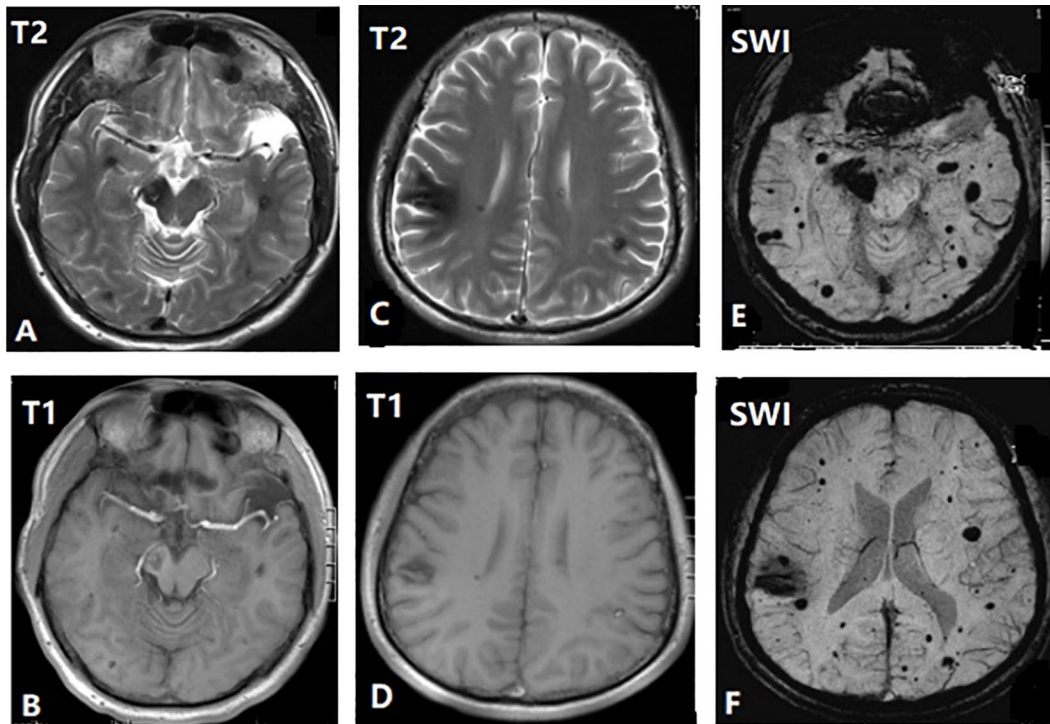


Figure 2. Brain MRI of the proband. The lesion of the cerebral peduncle is hyper-intense in the center and surrounded by a hypo-intense rim on T2-weighted imaging (a), and on T1-weighted imaging (b). The lesions of the cerebral cortex show mixed imaging on T2 and T1-weighted imaging (c, d). Brain lesions are more prominent on SWI, which shows multiple hypo-intense lesions (e, f). MRI, magnetic resonance imaging; SWI, susceptibility weighted imaging.

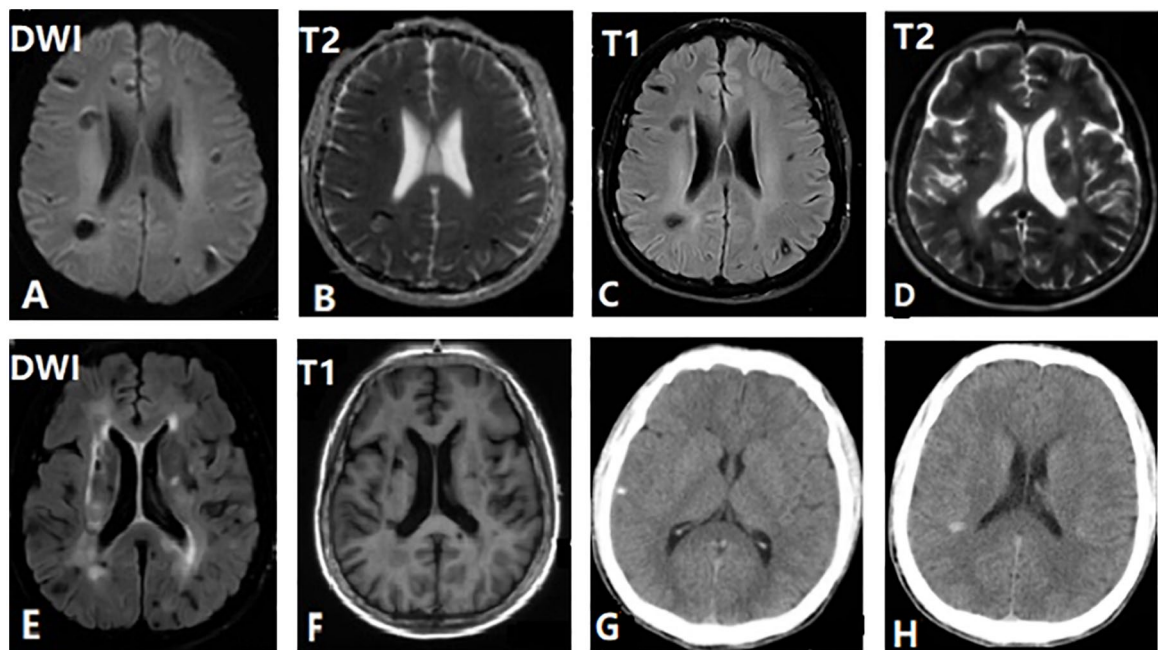


Figure 3. Brain MRI scans of the proband's older brother (a–c) and his mother (d–f) show multiple lesions in the cerebral cortex and centrum semiovale. These lesions are hypo-intense or mixed on T2/T1-weighted imaging, and more prominent on DWI. Brain CT of the proband's younger brother shows multiple high-density shadows (g, h). CT, computed tomography; DWI, diffusion-weighted imaging; MRI, magnetic resonance imaging; SWI, susceptibility weighted imaging.

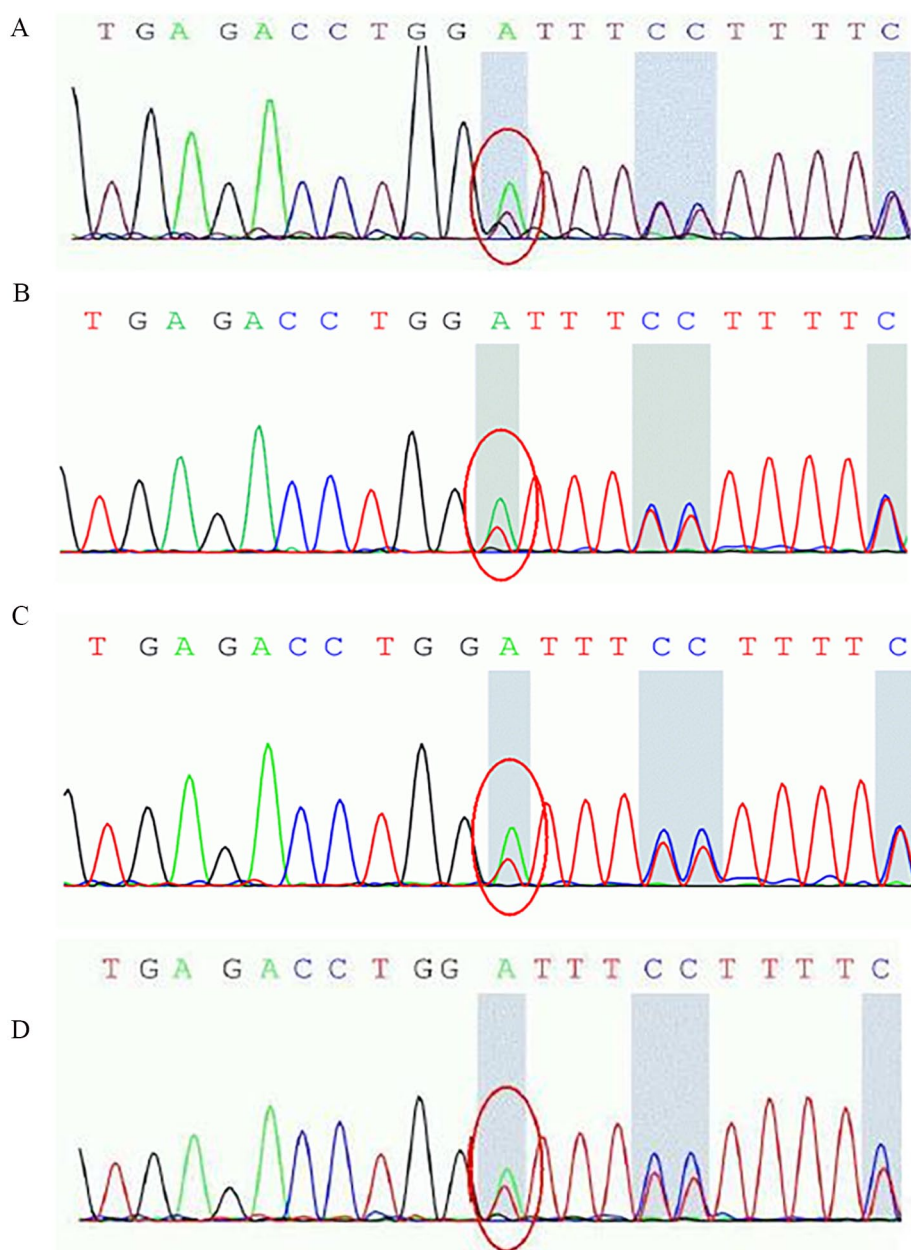


Figure 4. DNA sequencing analysis of the proband (a) showing the heterozygous deletion in the *PDCD10/CCM3* gene: c.165delT, p.N55fs (red circle). The same mutation was found in his brothers and mother (b–d).

prematurely terminated translation of the protein. The same mutation in the *CCM3* gene was found in the proband's brothers and mother, indicating that the mutation is associated with the disease in the family, and has not been reported before.

Treatment

So far, no medical therapy has been proven to be useful for CCM, especially for small multiple

lesions. Symptomatic treatment, such as controlling blood pressure and relieving headache, is the treatment of choice for the proband and his relatives at present.

Discussion

CCM is a relatively rare and occult vascular disease of the central nervous system. The prevalence of all types of CCM is approximately 0.1–0.5%.^{1,2}

The familial form of CCM is associated with three genes: *KRIT1/CCM1*, *MGC4607/CCM2*, and *PDCD10/CCM3*, of which *CCM3* is the most recently discovered and has more severe disease manifestations compared with the other two.^{5,6} To date, there are ~20 *CCM3* mutations that can lead to CCM. According to previous reports, most of the indels occur in the N-terminal region of *CCM3* gene⁷; however, the etiology, pathogenesis, and treatment of CCM are still not well understood. In our case, we found a novel *CCM3* mutation associated with CCM that has not been reported before, which is potentially due to highly conservation of this site. The fact that all family members of the proband have different clinical manifestations, and have been suffering from the disease since an early age, is similar to a previous study reported by Kevin Whitehead and his team, wherein they state that patients with a *CCM3* mutation have a greater chance of spontaneous hemorrhage, which presents at a younger age.⁸ All clinical manifestations of the proband and his family members indicate that the *CCM3* mutation may lead to severe clinical symptoms and poor prognosis.

So far, several gene mutations have been found to be associated with CCM in different populations. Notably, most of these gene mutations are heterozygous and are either frameshift, nonsense, or splice site mutations that result in different forms of the protein and eventually lead to CCM.⁹ In our study, DNA sequence analysis of the proband and his relatives revealed the presence of a deletion of T at the 165th position in the coding region of the *CCM3* gene. This heterozygous deletion changes the nucleotide sequence and leads to a frameshift in the gene. Consequently, protein translation is terminated prematurely. The clinical evidence and family history investigation reveal that the gene mutation is the cause of the disease in this case. However, the exact pathogenic mechanism is still unclear. Recently, studies have shown that the *CCM3* gene may be associated with cell apoptosis, and is involved in the *CCM1/2* pathway that is associated with the disease.¹⁰ Products of the *CCM3* gene play major roles in angiogenesis and endothelial cell junctions in neural tissue. Loss-of-function mutations in the *CCM3* gene lead to extensive vascular abnormalities and increased permeability.¹¹ Another study showed that oxidative stress and inflammation may have an important impact on the progress of CCM.¹² Due to the limited

numbers of CCM carriers, more studies are required to investigate the problem.

With the development of imaging technology, diagnosis of CCM has become easier than previously. CCM usually cannot be noticed through angiography. MRI has near-perfect sensitivity and excellent specificity, and, therefore, is the best available option for the detection and characterization of CCM.¹³ Generally, the appearance of CCM lesions on MRI scans differs from case to case. A typical image is presented as a mixed signal on both T1- and T2-weighted imaging. On T2-weighted imaging, the lesions have a hypo-intense rim, resulting in a ‘popcorn-like’ appearance; however, physiologic senile iron deposition will also cause hypo-intense lesion on normal brain.^{11,14} In our study, the proband and his relatives had similar MRI manifestations; the brain lesions were partly hypo-intense on T1-weighted imaging, whereas they were significantly more intense on T2-weighted imaging. Other lesions were hyper-intense in the center, and surrounded by a hypo-intense rim on T1/T2-weighted imaging. The reason we did not observe typical images in our case may be due to whether or not the lesions are hemorrhagic, and the time of the hemorrhage. It is possible that the differences in manifestation are associated with the underlying gene mutation. The proband also underwent complete SWI, which showed more lesions, and these lesions were more prominent than those seen on conventional MRI. This confirms the idea that brain SWI can accurately recognize deoxyhemoglobin and hemosiderin, and, therefore, is advantageous for the detection of CCM lesions.¹³ The diameter of the CCM lesions ranges from a few millimeters to several centimeters, and new lesions appear at a rate of 0.2–0.4 lesions per patient-year. The accuracy of brain MRI makes it a suitable method for the evaluation of brain lesion development.¹⁵ With the development of imaging technologies, diagnosis of CCM will be quicker and more accurate.

The current treatment for CCMs is surgical resection, and no other medical therapies have been approved so far. Laboratory and preclinical studies have shown that statins are a potential therapy for CCM⁸; however, more studies need to be done in the future. Based on studies of the CCM mechanism, some drugs have been tested in animals. In our case, the proband was suggested to take statins and to control his blood pressure. Follow up on further curative effects will be required.

Conclusion

CCM is a rare vascular disease of the central nervous system. In this study, we identified a novel heterozygous deletion in the *CCM3* gene. This finding expands the *CCM* gene mutation profile, and will be beneficial for genetic counseling. However, further laboratory studies and clinical assessments are necessary to identify the pathogenesis of CCM and potential targets for therapy.

Ethics statement

Written informed consent for publication of the case report was obtained from the patient and his relatives.

Acknowledgements

The authors thank Editage for proofreading the manuscript.

Funding

The authors disclosed receipt of the following financial support for the research, authorship, and publication of this article: This work was supported by the National Natural Science Foundation of China (No, 81601088; No, 81974191).

Conflict of interest statement

The authors declare that there is no conflict of interest.

ORCID iD

Xiao-yu Jiang  <https://orcid.org/0000-0002-6553-3241>

References

- Kim J. Introduction to cerebral cavernous malformation: a brief review. *BMB Rep* 2016; 49: 255–262.
- Otten P, Pizzolato GP, Rilliet B, *et al.* 131 cases of cavernous angioma (cavernomas) of the CNS discovered by retrospective analysis of 24,535 autopsies. *Neurochirurgie* 1989; 35: 82–83.
- Labauge P, Laberge S, Brunereau L, *et al.* Hereditary cerebral cavernous angiomas: clinical and genetic features in 57 French families. *Lancet* 1998; 352: 1892–1897.
- Yu W, Jin H, You Q, *et al.* A novel PDCD10 gene mutation in cerebral cavernous malformations: a case report and review of the literature. *J Pain Res* 2019; 12: 1127–1132.
- Shenkar R, Shi C, Rebeiz T, *et al.* Exceptional aggressiveness of cerebral cavernous malformation disease associated with PDCD10 mutations. *Genet Med* 2015; 17: 188–196.
- Zhou HJ, Qin L, Zhang H, *et al.* Augmented endothelial exocytosis of angiopoietin-2 resulting from CCM3-deficiency contributes to the progression of cerebral cavernous malformation. *Nat Med* 2016; 22: 1033–1042.
- Kumar A, Bhandari A and Goswami C. Surveying genetic variants and molecular phylogeny of cerebral cavernous malformation gene, CCM3/PDCD10. *Biochem Biophys Res Commun* 2014; 455: 98–106.
- Akers A, AI-Shahi Salman R, Awad IA, *et al.* Synopsis of guidelines for the clinical management of cerebral cavernous malformations: consensus recommendations based on systematic literature review by the angioma alliance scientific advisory board clinical experts panel. *Neurosurgery* 2017; 80: 665–680.
- Spiegler S, Rath M, Paperlein C, *et al.* Cerebral cavernous malformations: an update on prevalence, molecular genetic analyses, and genetic counselling. *Mol Syndromol* 2018; 9: 60–69.
- Cigoli MS, Avemaria F, De Benedetti S, *et al.* PDCD10 gene mutations in multiple cerebral cavernous malformations. *PLoS One* 2014; 9: e110438.
- Mouchtouris N, Chalouhi N, Chitale A, *et al.* Management of cerebral cavernous malformations: from diagnosis to treatment. *ScientificWorldJournal* 2015; 2015: 808314.
- Retta SF and Glading AJ. Oxidative stress and inflammation in cerebral cavernous malformation disease pathogenesis: two sides of the same coin. *Int J Biochem Cell Biol* 2016; 12: 254–270.
- Campbell PG, Jabbar P, Yadla S, *et al.* Emerging clinical imaging techniques for cerebral cavernous malformations: a systematic review. *Neurosurg Focus* 2010; 29: E6.
- Hiwatashi A, Kinoshita T, Moritani T, *et al.* Hypointensity on diffusion-weighted MRI of the brain related to T2 shortening and susceptibility effects. *AJR Am J Roentgenol* 2003; 181: 1705–1709.
- Revencu N and Vikkula M. Cerebral cavernous malformation: new molecular and clinical insights. *J Med Genet* 2006; 43: 716–721.