COLD SPRING HARBOR
Molecular Case Studies

RAPID COMMUNICATION

Parkes Weber syndrome with lymphedema caused by a somatic *KRAS* variant

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Abstract Parkes Weber syndrome is a vascular malformation overgrowth condition typically involving the legs. Its main features are diffuse arteriovenous fistulas and enlargement of the limb. The condition has been associated with pathogenic germline variants in *RASA1* and *EPHB4*. We report two individuals with Parkes Weber syndrome of the leg and primary lymphedema containing a somatic *KRAS* variant (NM_004985.5:c.35G > A; p.Gly12Asp). *KRAS* variants, which cause somatic intracranial and extracranial arteriovenous malformations, also result in Parkes Weber syndrome with lymphatic malformations.

INTRODUCTION

Parkes Weber syndrome is a fast-flow vascular malformation overgrowth condition (Revencu et al. 2008). Its main features consist of diffuse arteriovenous fistulas throughout an extremity, capillary malformation, and limb overgrowth. Arteriovenous shunting can cause a warm limb and congestive heart failure. Treatment includes embolization, excision of overgrown soft tissues, and amputation. Two causes of Parkes Weber syndrome have been identified: germline variants in *RASA1* (CM-AVM1) (Revencu et al. 2008) and germline variants in *EPHB4* (CM-AVM2) (Amyere et al. 2017). Recently a somatic variant in *RASA1* has been shown to cause the condition as well (Flores Daboub et al. 2020).

CASE PRESENTATION

Two unrelated male patients referred to our Vascular Anomalies Center were diagnosed with Parkes Weber syndrome by physical examination, magnetic resonance imaging (MRI), and angiography (Fig. 1). Both subjects had significant left leg enlargement, edema, lymphorrhea, faint cutaneous vascular stains, and repeated infections requiring hospitalization and antibiotic therapy. Neither individual had baseline cardiac overload or a family history of capillary or arteriovenous malformations. Imaging demonstrated diffuse arteriovenous fistulas throughout the leg, subcutaneous microcystic lymphatic anomalies, and lymphedema. Both individuals were negative for germline *RASA1* variants (data not shown) and had overgrown skin and subcutaneous tissue excised to reduce the size of their limb.

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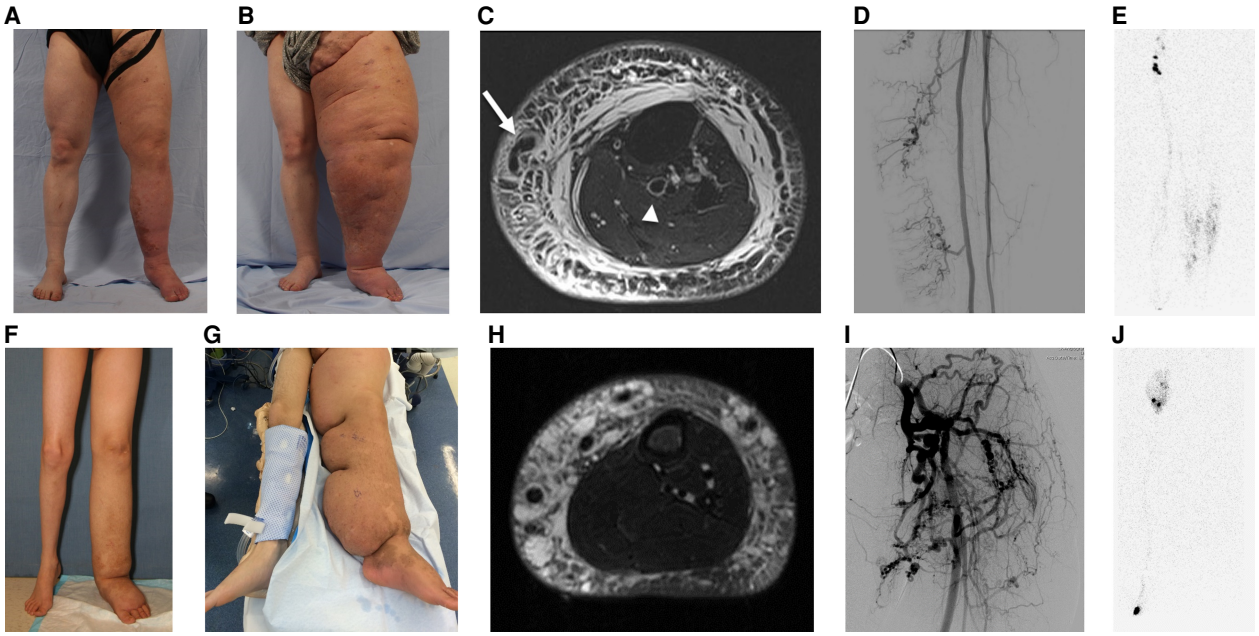

Figure 1. Parkes Weber syndrome with lymphedema caused by a somatic *KRAS* (NM_004985.5:c.35G > A; p.Gly12Asp) variant. (*A*) *Patient 1*: 34 yr-old male with left leg swelling and a capillary malformation of his posterior leg and buttock. (*B*) Significant progression of his disease at age 37 yr. (*C*) Axial T2-weighted fat-saturated magnetic resonance image of the left leg. Extensive reticular hyperintense signal within the overgrown subcutis. The posterior tibial vessels (arrowhead) are dilated with superficial enlarged medial veins (arrow). (*D*) Angiography shows a dilated posterior tibial artery with multiple enlarged, tortuous branches supplying the subcutaneous tissue of the medial aspect of the leg around the great saphenous vein. (*E*) Lymphoscintigram image 45 min following radiolabeled tracer injection into the feet demonstrates absence of flow to the inguinal nodes and dermal backflow confirming lymphedema. (*F*) *Patient 2*: 13-yr-old male with left leg overgrowth. (*G*) Enlargement of his affected extremity at age 22. (*H*) Magnetic resonance imaging (MRI) shows dilated vessels, subcutaneous microcystic lymphatic anomalies, and edema. (*I*) Angiogram exhibits arteriovenous connections. (*J*) Lymphoscintigraphy illustrates lymphedema of the left leg (no inguinal node tracer uptake 45 min after injection).

TECHNICAL ANALYSIS

The resected specimens underwent targeted exome sequencing with OncoPanel (Goss et al. 2019) (Brigham and Women's Hospital Department of Pathology). The OncoPanel assay surveys exonic DNA sequences of 447 cancer genes and 191 regions across 60 genes for rearrangement detection. DNA is isolated from tissue containing at least 20% tumor nuclei and analyzed by massively parallel sequencing using a solution-phase Agilent SureSelect hybrid capture kit and an Illumina HiSeq 2500 sequencer. The specimen for Patient 1 generated 13,848,235 aligned, high-quality reads with a mean of 302 reads across all targeted exons and 98% of all exons having more than 30 reads. The specimen for Patient 2 generated 8,119,343 aligned, high-quality reads with a mean of 233 reads across all targeted exons and 97% of all exons having more than 30 reads.

RESULTS

Both patient specimens contained a mosaic *KRAS* variant (NM_004985.5:c.35G > A; p.Gly12Asp) (Table 1). Variant allele fractions were 13% (Patient 1) and 9% (Patient 2).

Table 1. Variant table

Gene	Genomic location	HGVS cDNA	HGVS protein	Zygosity	Parent of origin	Variant interpretation
KRAS	Chr 12: 25245350 (GRCh38)	NM_004985.5: c.35G > A	p.Gly12Asp	Somatic heterozygous	N/A	Pathogenic

Droplet digital polymerase chain reaction (PCR) confirmed the variant in the resected tissue of Patient 1 (Patient 2 was not tested) and did not identify the variant in the whole blood DNA of either subject. Testing for *EPHB4* variants was not performed.

DISCUSSION

This report shows that *KRAS* is a locus for Parkes Weber syndrome. Similar to Parkes Weber syndrome caused by germline *RASA1* and *EPHB4* variants, the individuals had arteriovenous shunting and extremity overgrowth. In contrast, the patients exhibited lymphedema, lymphorrhea, repeated infections, lighter-colored capillary malformations, and significant enlargement of the affected extremity. The same somatic *KRAS* variant (NM_004985.5:c.35G > A; p.Gly12Asp) recently was reported in a 3-yr-old male with lower extremity overgrowth, arteriovenous fistulas, and capillary malformation; lymphatic dysfunction was not present (Schmidt et al. 2021).

Pathogenic *KRAS* variants are the most common cause for intracranial arteriovenous malformations (Nikolaev et al. 2018), and most extracranial arteriovenous malformations result from *MAP2K1* variants (Couto et al. 2017). Extracranial arteriovenous malformations with *KRAS* variants are associated with intramuscular fast-flow vascular anomaly (Goss et al. 2019) as well as lesions resembling congenital hemangiomas (Sudduth et al. 2020). Because *KRAS* variants cause intramuscular fast-flow vascular anomaly, its association with Parkes Weber syndrome, which contains diffuse intramuscular arteriovenous fistulas, is consistent. The association of the *KRAS* variant with primary lymphedema also has precedent because lymphatic abnormalities can occur in *KRAS*-related cardiofaciocutaneous and Noonan syndromes (Schubbert et al. 2006; Morcaldi et al. 2015).

Somatic variants in *KRAS* also have been associated with encephalocraniocutaneous lipomatosis and Schimmelpenning syndrome (Groesser et al. 2012; McDonell et al. 2018). Patients with Schimmelpenning syndrome have an increased risk of vascular anomalies, including lymphatic malformations (Greene et al. 2007). Neither of the patients we describe in this report had clinical findings diagnostic for encephalocraniocutaneous lipomatosis or Schimmelpenning syndrome (e.g., nevus psiloliparus, choristomas, macrocephaly, nevus sebaceous, hypoplastic bones, ocular abnormalities).

This report confirms that Parkes Weber syndrome can be caused by a mosaic *KRAS* variant. Although Parkes Weber syndrome resulting from *RASA1* or *EPHB4* germline variants has an overlapping phenotype, *KRAS*-related Parkes Weber syndrome can also include lymphedema. Individuals with suspected Parkes Weber syndrome without *RASA1* or *EPHB4* germline variants should be tested for somatic *KRAS* variants, especially if they exhibit lymphatic malformations. Pharmacotherapy against *KRAS* or other targets in this pathway might prevent worsening extremity overgrowth.

ADDITIONAL INFORMATION

Database Deposition and Access

The generated data set has been deposited in the ClinVar database (https://www.ncbi.nlm.nih.gov/clinvar/) under accession number SCV001739511.

Ethics Statement

The Committee on Clinical Investigation at Boston Children's Hospital approved this study. All procedures performed were in accordance with the ethical standards of the institutional and national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

Author Contributions

W.E. curated the data, administered the project, visualized and conceptualized the project, reviewed and edited the writing, and provided formal analysis, methodology, validation, and investigation; C.L.S., D.J.K., and P.J.S. curated the data, administered the project, visualized the project, administered the software, reviewed and edited the writing, and provided formal analysis, methodology, validation, and investigation; A.H.T. and S.J.F. visualized the project, reviewed and edited the writing, and provided resources, supervision, and validation; A.A. curated the data, administered the software, visualized the project, reviewed and edited the writing, and provided formal analysis, methodology, and validation; D.M.A. curated the data, administered the software, visualized the project, reviewed and edited the writing, established the methodology, and provided resources, supervision, and validation; and A.K.G. curated the data, administered the project, administered the software, visualized the project, wrote the original draft, reviewed and edited the writing, conceptualized the project, and provided formal analysis, funding acquisition, methodology, resources, supervision, validation, and investigation.

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Competing Interest Statement

The authors have declared no competing interest.

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REFERENCES

Amyere M, Revencu N, Helaers R, Pairet E, Baselga E, Cordisco M, Chung W, Dubois J, Lacour J, Martorell L, et al. 2017. Germline loss-of-function mutations in EPHB4 cause a second form of capillary malformation-arteriovenous malformation (CM-AVM2) deregulating RAS-MAPK signaling. *Circulation* **136:** 1037–1048. doi:10.1161/CIRCULATIONAHA.116.026886

Couto JA, Huang AY, Konczyk DJ, Goss JA, Fishman SJ, Mulliken JB, Warman ML, Greene AK. 2017. Somatic *MAP2K1* mutations are associated with extracranial arteriovenous malformation. *Am J Hum Genet* **100:** 546–554. doi:10.1016/j.ajhg.2017.01.018

Flores Daboub JA, Grimmer JF, Frigerio A, Wooderchak-Donahue W, Arnold R, Szymanski J, Longo N, Bayrak-Toydemir P. 2020. Parkes Weber syndrome associated with two somatic pathogenic variants in *RASA1*. *Cold Spring Harb Mol Case Stud* **6:** a005256. doi:10.1101/mcs.a005256

Goss JA, Konczyk DJ, Smits PJ, Kozakewich HPW, Alomari AI, Al-Ibraheemi A, Taghinia AH, Dickie BH, Adams DM, Fishman SJ, et al. 2019. Intramuscular fast-flow vascular anomaly contains somatic *MAP2K1* and *KRAS* mutations. *Angiogenesis* **22:** 547–552. doi:10.1007/s10456-019-09678-w

Greene AK, Rogers GF, Mulliken JB. 2007. Schimmelpenning syndrome: an association with vascular anomalies. *Cleft Palate Craniofac J* **44:** 208–215. doi:10.1597/06-025.1

Groesser L, Herschberger E, Ruetten A, Ruivenkamp C, Lopriore E, Zutt M, Langmann T, Singer S, Klingseisen L, Schneider-Brachert W, et al. 2012. Postzygotic *HRAS* and *KRAS* mutations cause nevus sebaceous and Schimmelpenning syndrome. *Nat Genet* **44:** 783–787. doi:10.1038/ng.2316

McDonell LM, Leung GKC, Daoud H, Ip J, Chim S, Luk HM, Lan L, Boycott KM, Chung BHY. 2018. Mosaic *KRAS* mutation in a patient with encephalocraniocutaneous lipomatosis and renovascular hypertension. *Am J Med Genet A* **176:** 2523–2527. doi:10.1002/ajmg.a.40349

Morcaldi G, Bellini T, Rossi C, Maghnie M, Boccardo F, Bonioli E, Bellini C. 2015. Lymphodysplasia and KRAS mutation: a case report and literature review. *Lymphology* **48:** 121–127.

Nikolaev SI, Vetiska S, Bonilla X, Boudreau E, Jauhiainen S, Rezai Jahromi B, Khyzha N, DiStefano PV, Suutarinen S, Kiehl TR, et al. 2018. Somatic activating *KRAS* mutations in arteriovenous malformations of the brain. *N Engl J Med* **378:** 250–261. doi:10.1056/NEJMoa1709449

Revencu N, Boon LM, Mulliken JB, Enjolras O, Cordisco MR, Burrows PE, Clapuyt P, Hammer F, Dubois J, Baselga E, et al. 2008. Parkes Weber syndrome, vein of Galen aneurysmal malformation, and other fast-flow vascular anomalies are caused by *RASA1* mutations. *Hum Mutat* **29:** 959–965. doi:10.1002/humu.20746

Schmidt VF, Wieland I, Wohlgemuth WA, Ricke J, Wildgruber M, Zenker M. 2021. Mosaic RASopathy due to *KRAS* variant G12D with segmental overgrowth and associated peripheral vascular malformations. *Am J Med Genet A* **38:** 331–336.

Schubbert S, Zenker M, Rowe SL, Böll S, Klein C, Bollag G, van der Burgt I, Musante L, Kalscheuer V, Wehner LE, et al. 2006. Germline *KRAS* mutations cause Noonan syndrome. *Nat Genet* **38:** 331–336. doi:10.1038/ng1748

Sudduth CL, McGuire AM, Smits PJ, Konczyk DJ, Al-Ibraheemi A, Fishman SJ, Greene AK. 2020. Arteriovenous malformation phenotype resembling congenital hemangioma contains *KRAS* mutations. *Clin Genet* **98:** 595–597. doi:10.1111/cge.13833