Patient report **Open Access**

Shira Harel, Ana S.A. Cohen, Khalid Hussain, Sarah E. Flanagan, Kamilla Schlade-Bartusiak, Millan Patel, Jaques Courtade, Jenny B.W. Li, Clara Van Karnebeek, Harley Kurata, Sian Ellard, Jean-Pierre Chanoine and William T. Gibson*

Alternating hypoglycemia and hyperglycemia in a toddler with a homozygous p.R1419H *ABCC8* mutation: an unusual clinical picture

Abstract

Background: Inheritance of two pathogenic *ABCC8* alleles typically causes severe congenital hyperinsulinism. We describe a girl and her father, both homozygous for the same *ABCC8* mutation, who presented with unusual phenotypes.

Methods: Single nucleotide polymorphism microarray and Sanger sequencing were performed. Western blot, rubidium efflux, and patch clamp recordings interrogated the expression and activity of the mutant protein.

Results: A 16-month-old girl of consanguineous descent manifested hypoglycemia. She had dysregulation of insulin secretion, with postprandial hyperglycemia followed by hypoglycemia. Microarray revealed homozygosity for the regions encompassing *KCNJ11* and *ABCC8*. Her father had developed diabetes at 28 years of age. Sequencing of *ABCC8* identified a homozygous missense mutation, p.R1419H, in both individuals. Functional studies showed absence of working K_{ATP} channels.

Conclusion: This is the first description of a homozygous p.R1419H mutation. Our findings highlight that homozygous loss-of-function mutations of *ABCC8* do not necessarily translate into early-onset severe hyperinsulinemia.

Keywords: *ABCC8*; diabetes; hyperglycemia; hyperinsulinism; hypoglycemia.

DOI 10.1515/jpem-2014-0265
Received June 20, 2014; accepted October 30, 2014; previously published online January 20, 2015

***Corresponding author: William T. Gibson,** Child and Family Research Institute, Department of Medical Genetics, British Columbia Children's Hospital, 950 West 28th Avenue, Vancouver, BC, V5Z 4H4, Canada, Phone: +1-604-875-2000 ext. 5523, Fax: +1-604-875-2373, E-mail: wtgibson@cfri.ubc.ca
Shira Harel and Jean-Pierre Chanoine: Endocrinology and Diabetes Unit, Department of Pediatrics, British Columbia Children's Hospital, University of British Columbia, Vancouver, Canada
Ana S.A. Cohen, Millan Patel and William T. Gibson: Department of Medical Genetics, University of British Columbia, Vancouver, Canada; and Child and Family Research Institute, Vancouver, Canada
Khalid Hussain: Institute of Biomedical and Clinical Science, University of Exeter Medical School, Exeter, UK; and Developmental Endocrinology Research Group, Clinical and Molecular Genetics, Institute of Child Health, University College London, London, UK
Sarah E. Flanagan and Sian Ellard: Institute of Biomedical and Clinical Science, University of Exeter Medical School, Exeter, UK
Kamilla Schlade-Bartusiak: Child and Family Research Institute, Vancouver, Canada; and Department of Pathology and Laboratory Medicine, University of British Columbia, Vancouver, Canada
Jaques Courtade: Child and Family Research Institute, Vancouver, Canada
Jenny B.W. Li and Harley Kurata: Department of Anesthesiology, Pharmacology and Therapeutics, University of British Columbia, Vancouver, Canada
Clara Van Karnebeek: Division of Biochemical Diseases, Department of Pediatrics, British Columbia Children's Hospital, University of British Columbia, Vancouver, Canada

Introduction

Autosomal recessive inheritance of two abnormal *ABCC8* or *KCNJ11* alleles causes severe forms of hyperinsulinism (HI) (1–4). Encoded by two adjacent genes on chromosome 11p, the sulfonylurea receptor (SUR1) modulates channel activity, whereas the inwardly rectifying potassium channel (Kir6.2) makes up the ion-conducting pore of the hetero-octameric ATP-sensitive potassium channel (K_{ATP}) (5, 6). The most common outcome of recessively inherited *ABCC8* gene mutations is to prevent expression of functional channels, leading to severe HI that presents early in life and is unresponsive to diazoxide. Most patients require subtotal pancreatectomy (7, 8). In some

cases, as reported for a splice site mutation of *ABCC8*, there can be variability in the age and severity of presentation in homozygous individuals (9, 10).

The natural history of patients who do not require pancreatectomy can include remission of the hypoglycemia, with progression to diabetes in early adulthood (11). However, long-term data is limited. These patients have been observed to have both hypoglycemic and hyperglycemic episodes a few years after presentation and pathologic responses to the oral glucose tolerance test have been described after 10 years of age (11). A blunted insulin response to intravenous dextrose was reported, suggesting reduced glucose sensitivity of the SUR-deficient β-cell (12), a condition referred to colloquially as "glucose-blindness".

In this report, we describe the unusual clinical course and workup of a patient who is homozygous for a p.R1419H mutation in *ABCC8*, a seemingly severe coding mutation that prevents maturation of K_{ATP} channels to the cell surface. The patient's presentation is a rare example of a weak phenotype with relatively late onset, inconsistent with the expected severity of the mutation at the cellular level.

Materials and methods

Clinical data

Clinical information was obtained by chart review, with approval from the Clinical Research Ethics Board that covers British Columbia Children's Hospital.

DNA mutation analysis

Affymetrix CytoScanHD (Affymetrix, Inc., Santa Clara, CA, USA) single nucleotide polymorphism (SNP) array analysis was done according to the manufacturer's protocol at the Cytogenetics Laboratory at British Columbia Children's Hospital. SNP marker results were mapped to the February 2009 build of the human genome (GRCh37/hg19). Sanger sequencing of the *KCNJ11* and *ABCC8* genes was performed as previously described (13).

Non-radioactive rubidium efflux assay

Rb^+ efflux assays were carried out on CosM6 cells transfected with Kir6.2 and SUR1 (WT or SUR1[R1419H] mutant) as previously described (14). Rb^+ concentrations were determined by flame atomic absorption spectroscopy using an Aurora ICR8000 instrument (Aurora, Vancouver, Canada).

Patch clamp electrophysiology

Patch-clamp experiments of CosM6 cells transfected with Kir6.2 and SUR1 were carried out at room temperature, using a perfusion chamber equipped for rapid solution exchange apparatus, as previously described (15).

Western blots

CosM6 cells transfected with combinations of Kir6.2, SUR1, and SUR1[R1419H] were lysed with RIPA buffer at 4°C, and lysates were clarified by centrifugation. Protein concentration of lysates was determined with a BCA assay kit (Pierce, Rockford, IL, USA) and equal amounts of lysate were separated by SDS-PAGE (7.5% gel). Blots were probed with a monoclonal anti-SUR1 antibody (UC Davis/NIH NeuroMab Facility, Davis, CA, USA) and visualized with a Femto ECL reagent (Pierce) using a FluorChemSP (Alpha Innotech, San Leandro, CA, USA). Spot densitometry was carried out using AlphaEase software (Alpha Innotech).

Results

A previously healthy 16-month-old girl presented with hypoglycemia of 1.2 mmol/L (22 mg/dL) in the context of lethargy during a gastrointestinal infection. Retrospectively, the parents recalled episodes of irritability over the previous 4 months, including episodes of medial eye deviation that could represent hypoglycemic seizures. She had been born at term, weighing 3490 g, after a pregnancy complicated by gestational diabetes, with no documented history of neonatal hypoglycemia. She grew along the 50th percentiles for length and weight. She was still breastfeeding frequently. She had normal motor milestones with mild speech delay.

The patient's father reported having seizures as an infant in Pakistan. He could not recall whether they were febrile seizures or otherwise associated with intercurrent illness. He had normal intellect and he was diagnosed with diabetes at 28 years of age. His body mass index (BMI) was 22.4 kg/m^2 (−0.21 standard deviation) at that time. Medical records from the time of diagnosis were unavailable. He was managed by his family physician according to common guidelines with glyburide and metformin, but had not previously been tested genetically. HbA1c at 45 years of age was 8.1% (normal 4.5–6). The patient's mother had diet-controlled gestational diabetes. Her BMI was 21.6 kg/m^2 (−0.04 standard deviation). The parents were first cousins through their mothers and second cousins through their fathers, and each of their pairs of parents were also related to each other (Figure 1).

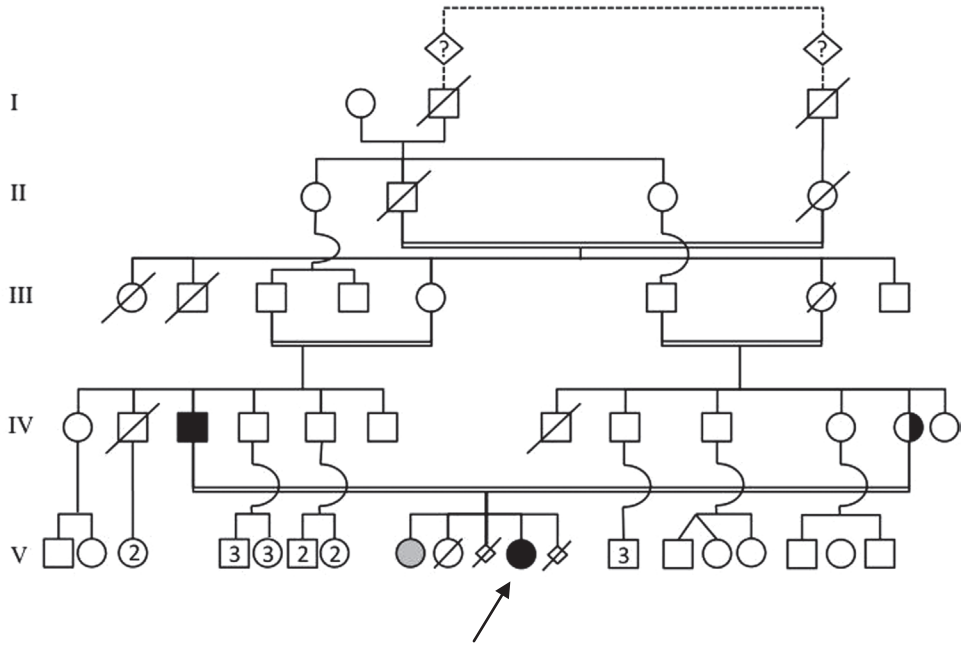

Figure 1 Pedigree chart. Pedigree chart showing the proband (V-18, arrow), her father (IV-3), and mother (IV-11). The family is of Pakistani descent. Her sister is shown in gray because she is asymptomatic and has yet to have genetic testing. Another sister died in an accident. The patient's mother had two miscarriages.

The patient had recurrent hypoglycemia in hospital and critical samples demonstrated alternating ketotic and non-ketotic hypoglycemia, with detectable insulin levels. An extensive workup was otherwise normal (Tables 1 and 2). Aberrant insulin processing was ruled out as a cause (Supplementary Table 1). Finally, a diagnosis of HI was entertained based on the insulin levels, associated with relatively low β-hydroxybutyrate, as well as a glycemic response to glucagon [increase of 3.4 mmol/L (61 mg/dL)]. Monitoring after a 2 g/kg carbohydrate meal showed postprandial hyperglycemia, 10.8 mmol/L (193 mg/dL) with relative hypoinsulinemia, 98 pmol/L (16.3 μIU/mL), followed by hypoglycemia, 2.1 mmol/L (38 mg/dL) with relative hyperinsulinemia, 34 pmol/L (5.6 μIU/mL) after 4.5 h (Figure 2), confirming inappropriate regulation of insulin secretion.

The patient did not respond to diazoxide and it was stopped at a dose of 12 mg/kg/day due to severe side effects. She also did not respond to boluses of up to 5 μg/kg of octreotide. She was eventually placed on a continuous subcutaneous glucagon infusion (0.7 mg/day) with a significant improvement in blood glucose readings. She had a gastrostomy tube placed with daytime bolus feeds and continuous night-time feeding of Pediasure (Abbott Laboratories, Abbott Park, IL, USA). The glucagon infusion was decreased gradually and stopped altogether after 5 months. She remains on continuous night-time feeding and eats orally frequently during the day. With this regimen, she experiences mild hypoglycemias, as well as postprandial hyperglycemias up to 12.5 mmol/L (225 mg/dL) every 2–3 days.

Table 1 Critical samples during hypoglycemia.

Test	I	II	III	IV
Glucose, mmol/L	2.0	2.3	1.7	2.0
β-Hydroxybutyrate, mmol/L	0.93	0.81	<0.1	0.1
Insulin, pmol/L	20	10	30	24
Cortisol, nmol/L	216[a]			
Growth hormone, μg/L	0.42[a]			

In the context of hypoglycemia, normal β-hydroxybutyrate >0.3 mmol/L, insulin <10 pmol/L, cortisol >500 nmol/L, and growth hormone >5.6 μg/L. [a]Following these initial results, stimulation tests for cortisol and growth hormone were performed with normal results, as per Table 2.

Microarray studies (Supplementary Table 2) revealed homozygosity for multiple genomic regions, consistent with the family history of consanguinity. The region surrounding *KCNJ11* and *ABCC8* was identified as the best candidate region for harboring rare variants with major effects on the phenotype. *KCNJ11* and *ABCC8* mutation analysis was performed at the University of Exeter Medical School, Exeter, UK, and identified a homozygous missense coding mutation, p.R1419H (c.4256G>A) in the *ABCC8* gene. Sanger sequencing of the patient and her parents (Supplementary Figure 1A–C) found that the mother was heterozygous and the father homozygous for the same mutation.

Table 2 Metabolic workup of the proband.

Test	Value	Normal range
Lactate, mmol/L	1.2	0.5–2.2
Ammonia, µmol/L	<9	9–33
Cortisol secretion in response to ACTH test[a], nmol/L	891	>500
Growth hormone secretion in response to glucagon test[b], µg/L	5.95	>5.6
ALT, U/L	27	5–45
Uric acid, µmol/L	175	105–300
CPK, U/L	71	60–305
Serum free fatty acids (glucose: 2.3 mmol/L), µmol/L	709	100–900
Lipid profile	Normal	
Plasma amino acid profile	Unremarkable	
Bloodspot and serum acylcarnitine profile	Unremarkable	
Urine organic acid profile	Unremarkable	
Iso-electric focusing transferrins	Unremarkable	
Newborn metabolic screening[c]	Negative	
Urine sulfonylurea level	Undetectable	
Glycogen storage disorders panel[d] (GSD0, 1, 2, 3, 4, 6, 9)	Normal	

[a]Standard protocol low dose adrenocorticotropic hormone (ACTH) test 1 µg. [b]Standard protocol growth hormone stimulation test with glucagon, 0.03 mg/kg. [c]Newborn metabolic screening in British Columbia targets amino acid disorders, fatty acid oxidation disorders, organic acid disorders, galactosemia, congenital hypothyroidism, hemoglobinopathies, and cystic fibrosis. [d]Performed by massively parallel sequencing (BCM-MitomeNGS[SM]).

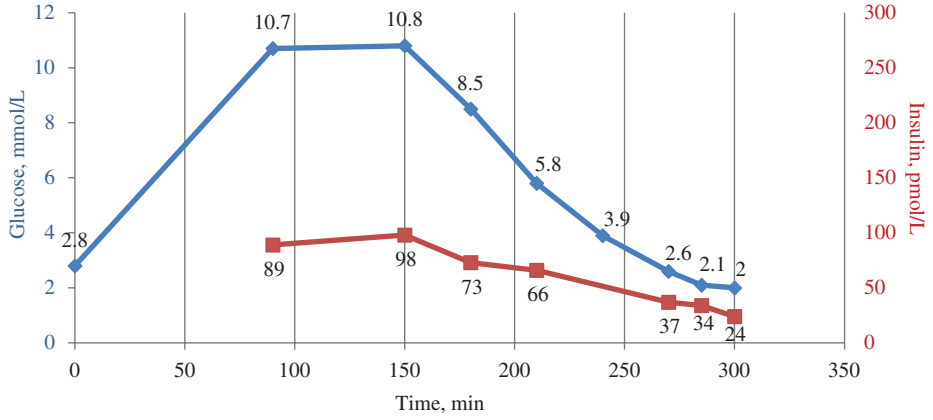

Figure 2 The patient's post-prandial glucose and insulin levels. Monitoring after a 2 g/kg carbohydrate meal showed post-prandial hyperglycemia (10.8 mmol/L, normal <7.8 mmol/L) with relative hypoinsulinemia (98 pmol/L, normal post-prandial insulin 120–300 pmol/L), followed by hypoglycemia (2.1 mmol/L) with relative hyperinsulinemia (insulin 34 pmol/L, normal for hypoglycemia <10 pmol/L).

Previous functional data in the literature has suggested that the SUR1[R1419H] mutation prevents channels from reaching the cell surface (16). However, the relatively mild phenotypes of the patient and her father contrasted with this "classical" clinical picture of homozygous recessive *ABCC8* mutations, which are typically very severe and present early in life (7, 17, 18). Therefore, we tested the effects of this mutation in a reconstituted system, using Rb^+ efflux assays of CosM6 cells transfected with Kir6.2/KCNJ11 and WT or SUR1[R1419H] to measure functional K_{ATP} channel expression (Figure 3). Under conditions of metabolic inhibition (1 mM deoxyglucose+100 µM oligomycin), to maximally activate K_{ATP} channels, cells transfected with the SUR1[R1419H] variant (+Kir6.2) exhibited no Rb^+ efflux above untransfected cells, consistent with an absence of functional K_{ATP} channels at the cell surface (Figure 3A). Cells transfected with a 1:1 mixture of WT: SUR1[R1419H] to simulate a heterozygous state exhibited Rb^+ efflux activity intermediate between the WT-only and R1419H-only SUR1 cells. We examined the effects of SUR1[R1419H] subunits on cell surface maturation of K_{ATP} using the differential migration of mature and immature glycosylated forms of SUR1 on SDS-PAGE gels (19–21) (Figure 3B and C). Cells transfected with SUR1 alone exhibit a single molecular

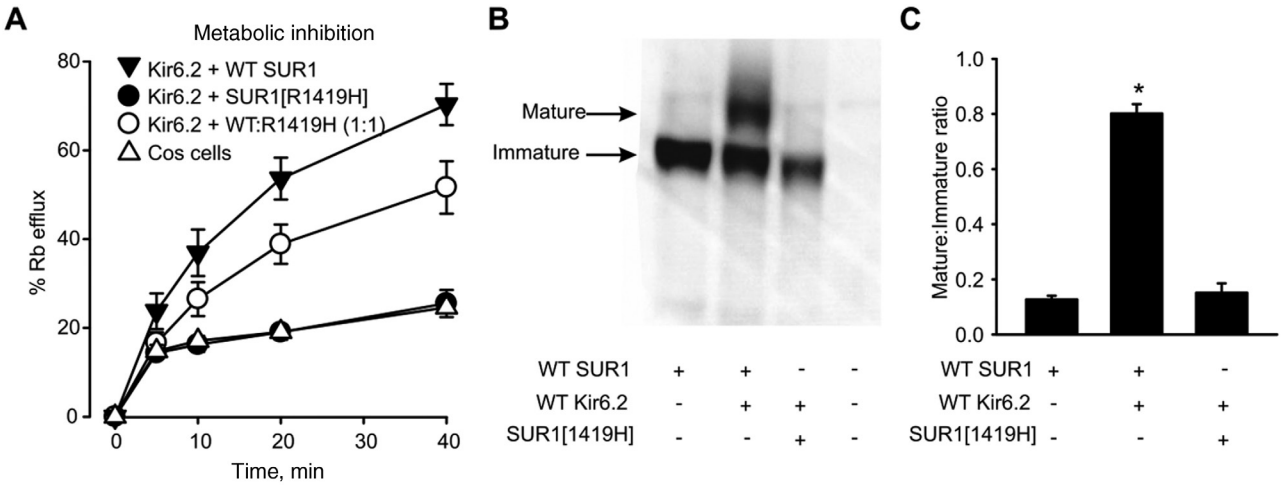

Figure 3 K_{ATP} channels containing SUR1[R1419H] exhibit loss-of-function due to poor trafficking to the cell surface. (A) Rb^+ efflux assays were performed in CosM6 cells transfected with the indicated plasmids in conditions of metabolic inhibition. K_{ATP} channels comprising SUR1[R1419H] to recapitulate the homozygous condition exhibit no Rb^+ efflux above untransfected cells. (B) Cell lysates from CosM6 cells transfected with indicated combinations of Kir6.2, SUR1, and SUR1[R1419H] were separated on SDS-PAGE and probed with an anti-SUR1 antibody (NeuroMab). (C) Spot densitometry was used to determine the relative abundance of mature vs. immature forms of SUR1 in various experimental conditions (n=5 matched experiments). No statistical significance in the mature:immature ratio was observed between cells transfected with SUR1 alone and Kir6.2+SUR1[R1419H].

weight band attributed to an immature form of the protein that fails to reach the plasma membrane. When co-transfected with Kir6.2 channels, a mature glycosylated band is apparent. However, co-transfection of SUR1[R1419H] with WT Kir6.2 channel subunits yields little or none of the mature glycosylated form (not statistically different from SUR1 alone; Figure 3C), consistent with a severe trafficking defect of this channel mutant. We also considered the possibility that the SUR1[R1419H] mutation could increase channel activity and partly counteract the trafficking defects, this might account for the relatively mild phenotype of the patient and her father if small numbers of channels containing the SUR1[R1419H] subunit could reach the cell surface. However, patch clamp recordings showed no significant change in nucleotide sensitivity of K_{ATP} channels containing the SUR1[R1419H] mutant (Supplementary Figure 2). Overall, the weak Rb^+ efflux and lack of protein maturation are consistent with trafficking defects that are apparent in many recessively inherited SUR1-mediated hyperinsulinemias (22–24). These data indicate that virtually no functional K_{ATP} channels are present in the plasma membrane of β-cells of the patient or her father.

Discussion

Compared to most reports of homozygous SUR1 mutations, our patient had late onset of symptoms and she exhibited post-prandial hyperglycemia, as well as fasting and rebound hypoglycemia already at presentation. Previous reports have suggested that the R1419H mutation causes a trafficking defect of the K_{ATP} complex (16), although the only affected patient yet reported was a G70E/R1419H compound heterozygote. That patient had a severe presentation at birth, requiring subtotal pancreatectomy. These authors concluded that the R1419H mutation was consistent with a "channel absent" molecular subclass common to recessive forms of HI (16, 23). Our patient and her father are the first humans to be described with this mutation in a homozygous state, and our in vitro studies confirmed the trafficking defect caused by the R1419H mutation in transfected CosM6 cells.

Mutations that prevent cell surface maturation of K_{ATP} channels typically lead to a very severe hyperinsulinemic phenotype in homozygotes. The relatively late and unusual clinical presentations of our patient and her father diverge significantly from this prediction. We uncovered one report of extreme phenotypic variability in a family with a homozygous *ABCC8* mutation (10), although it should be noted that this report involves a splicing site with unclear consequences on the generation of functional channels. In contrast, the R1419H is a coding mutation that appears to completely suppress expression of functional channels.

We were led to our top candidate genes by the clinical microarray that identified loss-of-heterozygosity (LOH) in

multiple chromosomal regions, including around *ABCC8* and *KCNJ11*. We began with a hypothesis that this was an autosomal recessive disease, based on parental consanguinity. We then used microarray analysis as an unbiased tool to create a list of candidate genes. We selected *ABCC8* and *KCNJ11* as plausible candidate genes based on the fact that there was LOH in the region around these two genes, and on the fact that we suspected an insulin secretion defect on clinical grounds.

Other glucose transporters and other "diabetes genes" for which the patient had LOH included *KLF11*, *SLC2A1*, *PCSK1*, and *TCF7L2* (Supplementary Table 2). Identification and validation of potential modifier loci was deemed unfeasible at this stage, because of the small number of affected individuals with the same *ABCC8* genotype available for study.

From a phenotypic perspective, the patient exhibited both a poor post-prandial insulin response and fasting hyperinsulinism, illustrating significant dysregulation of insulin secretion. Unfortunately, information on the father's phenotype over the years was not available, however, the history of unexplained seizures in infancy, and later diabetes in adulthood, can hint to his daughter's future phenotype. Information regarding insulin dysregulation in patients with recessive forms of SUR1 HI is sparse, mainly because most undergo subtotal pancreatectomy.

Co-existence of hypoglycemias and hyperglycemias has been reported only in long-term follow-up of non-pancreatectomized patients with diffuse SUR1 HI (11). In addition, Grimberg et al. (12) demonstrated the dysregulation of insulin secretion in patients with diffuse SUR1 HI. Lastly, the phenotype of $SUR1^{-/-}$ knockout mice (25) is surprisingly similar to our patient, as the mice do not have severe hypoglycemia and demonstrate a loss of first-phase and attenuated second-phase glucose-stimulated insulin secretion. Overall, the presence of K_{ATP} channel-independent pathways (26–28) may enable these patients and animal models to exhibit some sub-optimal glucose tolerance, although it remains unclear what factors underlie the relatively weak phenotype apparent in our patients. Nevertheless, our findings highlight that the inheritance of homozygous loss-of-function mutations of SUR1 does not necessarily translate into an early onset severe hyperinsulinemia. At some level, the fact that this family's phenotype does not match previously reported phenotypes is surprising, although perhaps it should not be, given the fact that broadening of genotype-phenotype correlations is a frequent outcome for families enrolled in the newer consortia-based genomic sequencing projects (29).

In conclusion, this the first description of a homozygous p.R1419H mutation of the ABCC8 gene. We were led to our candidate gene by looking at relevant regions of homozygosity on microarray. This case is particularly interesting because it diverges from the predicted severe phenotype that typically results from this "channel absent" subclass.

Acknowledgments: We wish to thank Dr. Charles Stanley (Children's Hospital of Philadelphia) for expert comments on the clinical care of our patient.

Conflict of interest statement: The authors have no relevant conflict of interest to disclose.

References

1. Thomas P, Ye Y, Lightner E. Mutation of the pancreatic islet inward rectifier Kir6.2 also leads to familial persistent hyperinsulinemic hypoglycemia of infancy. Hum Mol Genet 1996;5:1809–12.
2. Thomas PM, Cote GJ, Hallman DM, Mathew PM. Homozygosity mapping, to chromosome 11p, of the gene for familial persistent hyperinsulinemic hypoglycemia of infancy. Am J Hum Genet 1995;56:416–21.
3. Thomas PM, Cote GJ, Wohllk N, Haddad B, Mathew PM, et al. Mutations in the sulfonylurea receptor gene in familial persistent hyperinsulinemic hypoglycemia of infancy. Science 1995;268:426–9.
4. Thomas PM, Cote GJ, Wohllk N, Mathew PM, Gagel RF. The molecular basis for familial persistent hyperinsulinemic hypoglycemia of infancy. Proc Assoc Am Physicians 1996;108:14–9.
5. Inagaki N, Gonoi T, Clement JP 4th, Namba N, Inazawa J, et al. Reconstitution of IKATP: an inward rectifier subunit plus the sulfonylurea receptor. Science 1995;270:1166–70.
6. Aguilar-Bryan L, Nichols CG, Wechsler SW, Clement JP, Boyd AE, et al. Cloning of the beta cell high-affinity sulfonylurea receptor: a regulator of insulin secretion. Science 1995;268:423–6.
7. De León DD, Stanley CA. Mechanisms of disease: advances in diagnosis and treatment of hyperinsulinism in neonates. Nat Clin Pract Endocrinol Metab 2007;3:57–68.
8. Dunne MJ, Cosgrove KE, Shepherd RM, Aynsley-Green A, Lindley KJ. Hyperinsulinism in infancy: from basic science to clinical disease. Physiol Rev 2004;84:239–75.
9. De Lonlay P, Fournet JC, Touati G, Groos MS, Martin D, et al. Heterogeneity of persistent hyperinsulinaemic hypoglycaemia. A series of 175 cases. Eur J Pediatr 2002;161:37–48.
10. Kapoor RR, Flanagan SE, Ellard S, Hussain K. Congenital hyperinsulinism: marked clinical heterogeneity in siblings with identical mutations in the ABCC8 gene. Clin Endocrinol (Oxf) 2012;76:312–3.
11. Gussinyer M, Clemente M, Cebrian R, Yeste D, Albisu M, et al. Glucose intolerance and diabetes are observed in the long-term follow-up of nonpancreatectomized patients with persistent hyperinsulinemic hypoglycemia of infancy due to mutations in the ABCC8 gene. Diabetes Care 2008;31:1257–9.
12. Grimberg A, Ferry RJ Jr, Kelly A, Koo-McCoy S, Polonsky K, et al. Dysregulation of insulin secretion in children with congenital

hyperinsulinism due to sulfonylurea receptor mutations. Diabetes 2001;50:322–8.
13. Flanagan SE, Ellard S. Identification of mutations in the Kir6.2 subunit of the K(ATP) channel. Methods Mol Biol 2008;491:235–45.
14. Bruin JE, Erener S, Vela J, Hu X, Johnson JD, et al. Characterization of polyhormonal insulin-producing cells derived in vitro from human embryonic stem cells. Stem Cell Res 2014;12:194–208.
15. Khurana A, Shao ES, Kim RY, Vilin YY, Huang X, et al. Forced gating motions by a substituted titratable side chain at the bundle crossing of a potassium channel. J Biol Chem 2011;286:36686–93.
16. Tornovsky S, Crane A, Cosgrove KE, Hussain K, Lavie J, et al. Hyperinsulinism of infancy: novel ABCC8 and KCNJ11 mutations and evidence for additional locus heterogeneity. J Clin Endocrinol Metab 2004;89:6224–34.
17. Gloyn AL, Siddiqui J, Ellard S. Mutations in the genes encoding the pancreatic beta-cell KATP channel subunits Kir6.2 (KCNJ11) and SUR1 (ABCC8) in diabetes mellitus and hyperinsulinism. Hum Mutat 2006;27:220–31.
18. Dunne MJ, Kane C, Shepherd RM, Sanchez JA, James RF, et al. Familial persistent hyperinsulinemic hypoglycemia of infancy and mutations in the sulfonylurea receptor. N Engl J Med 1997;336:703–6.
19. Yan FF, Lin YW, MacMullen C, Ganguly A, Stanley CA, et al. Congenital hyperinsulinism associated ABCC8 mutations that cause defective trafficking of ATP-sensitive K+ channels: identification and rescue. Diabetes 2007;56:2339–48.
20. Schwappach B, Zerangue N, Jan YN, Jan LY. Molecular basis for K(ATP) assembly: transmembrane interactions mediate association of a K+ channel with an ABC transporter. Neuron 2000;26:155–67.
21. Zerangue N, Schwappach B, Jan YN, Jan LY. A new ER trafficking signal regulates the subunit stoichiometry of plasma membrane K(ATP) channels. Neuron 1999;22:537–48.
22. Shyng SL, Ferrigni T, Shepard JB, Nestorowicz A, Glaser B, et al. Functional analyses of novel mutations in the sulfonylurea receptor 1 associated with persistent hyperinsulinemic hypoglycemia of infancy. Diabetes 1998;47:1145–51.
23. Cartier EA, Conti LR, Vandenberg CA, Shyng SL. Defective trafficking and function of KATP channels caused by a sulfonylurea receptor 1 mutation associated with persistent hyperinsulinemic hypoglycemia of infancy. Proc Natl Acad Sci USA 2001;98:2882–7.
24. Taschenberger G, Mougey A, Shen S, Lester LB, LaFranchi S, et al. Identification of a familial hyperinsulinism-causing mutation in the sulfonylurea receptor 1 that prevents normal trafficking and function of KATP channels. J Biol Chem 2002;277:17139–46.
25. Seghers V, Nakazaki M, DeMayo F, Aguilar-Bryan L, Bryan J. Sur1 knockout mice. A model for K(ATP) channel-independent regulation of insulin secretion. J Biol Chem 2000;275:9270–7.
26. Sato Y, Aizawa T, Komatsu M, Okada N, Yamada T. Dual functional role of membrane depolarization/Ca2+ influx in rat pancreatic B-cell. Diabetes 1992;41:438–43.
27. Gembal M, Gilon P, Henquin JC. Evidence that glucose can control insulin release independently from its action on ATP-sensitive K+ channels in mouse B cells. J Clin Invest 1992;89:1288–95.
28. Straub SG, James RF, Dunne MJ, Sharp GW. Glucose activates both K(ATP) channel-dependent and K(ATP) channel-independent signaling pathways in human islets. Diabetes 1998;47:758–63.
29. Beaulieu CL, Majewski J, Schwartzentruber J, Samuels ME, Fernandez BA, et al. FORGE Canada Consortium: outcomes of a 2-year national rare-disease gene-discovery project. Am J Hum Genet 2014;94:809–17.

Supplemental Material: The online version of this article (DOI: 10.1515/jpem-2014-0265) offers supplementary material, available to authorized users.