

Congenital hyperinsulinism as the presenting feature of Kabuki syndrome: clinical and molecular characterization of 10 affected individuals

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Purpose: Describe the clinical and molecular findings of patients with Kabuki syndrome (KS) who present with hypoglycemia due to congenital hyperinsulinism (HI), and assess the incidence of KS in patients with HI.

Methods: We documented the clinical features and molecular diagnoses of 10 infants with persistent HI and KS via a combination of sequencing and copy-number profiling methodologies. Subsequently, we retrospectively evaluated 100 infants with HI lacking a genetic diagnosis, for causative variants in KS genes.

Results: Molecular diagnoses of KS were established by identification of pathogenic variants in *KMT2D* ($n = 5$) and *KDM6A* ($n = 5$). Among the 100 infants with HI of unknown genetic etiology, a KS diagnosis was uncovered in one patient.

Conclusions: The incidence of HI among patients with KS may be

higher than previously reported, and KS may account for as much as 1% of patients diagnosed with HI. As the recognition of dysmorphic features associated with KS is challenging in the neonatal period, we propose KS should be considered in the differential diagnosis of HI. Since HI in patients with KS is well managed medically, a timely recognition of hyperinsulinemic episodes will improve outcomes, and prevent aggravation of the preexisting mild to moderate intellectual disability in KS.

Genetics in Medicine (2019) 21:233–242; <https://doi.org/10.1038/s41436-018-0013-9>

Keywords: Hypoglycemia; hyperinsulinism; Kabuki syndrome; *KDM6A*; *KMT2D*

INTRODUCTION

Transient hypoglycemia is a common condition among neonates, typically resolving within the first few days of life. However, persistent neonatal hypoglycemia is a more significant finding that warrants further investigation.¹ One of the leading causes of persistent neonatal hypoglycemia is dysregulated insulin secretion by the pancreatic β -cells, termed congenital hyperinsulinism (HI). Affected neonates with the condition have inappropriately high levels of insulin at birth, causing recurrent episodes of hypoglycemia.² Timely

recognition of this condition is challenging, as the symptoms of hypoglycemia can be subtle in newborns.³ Hyperinsulinemic hypoglycemic episodes in neonates, if not recognized, are a leading cause of developmental delays and permanent neurologic damage.⁴ Pathogenic variants in 11 genes (*ABCC8*, *KCNJ11*, *GLUD1*, *GCK*, *HADH*, *UCP2*, *INSR*, *HNF1A*, *HNF4A*, *SLC16A1*, *PGM1*) have been reported as monogenic causes of nonsyndromic HI.^{5–14} The most common are inactivating pathogenic variants in the genes that encode for the two subunits of the pancreatic β -cell ATP-sensitive

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Submitted 20 November 2017; revised 20 November 2017; accepted: 20 March 2018

Published online: 15 June 2018

potassium channel, *ABCC8* and *KCNJ11*.^{5,6} Pathogenic variants in these two genes account for the majority of cases of HI, however approximately 50% of patients have an unknown genetic etiology.¹⁵

In addition to monogenic nonsyndromic causes of HI, several multisystemic genetic syndromes are associated with HI. The underlying molecular mechanisms that lead to HI in most of these syndromes are unclear. A significant proportion of patients with Beckwith–Wiedemann syndrome will present with HI in their childhood, whereas it has been noted as an infrequent occurrence in patients with Kabuki, Sotos, Costello, or Turner syndromes respectively.^{16–19}

Kabuki syndrome (KS) is a multiple congenital anomaly syndrome characterized by five cardinal manifestations: characteristic dysmorphic facial features, craniofacial/skeletal anomalies, dermatoglyphic abnormalities, mild to moderate intellectual disability and postnatal growth deficiencies.^{20,21} Patients with KS typically have characteristic dysmorphic facial features, including long palpebral fissures of the eye, eversion of the lower lateral eyelid, long and dense eyelashes, arched and broad eyebrows, depressed nasal tip, thin upper lip vermilion coupled with a full lower lip, and prominent ears. Skeletal and dermatoglyphic abnormalities are also commonly observed, and postnatal growth deficiencies may present as short stature and/or failure to thrive. Other findings that may be observed at a variable rate in KS include persistent hypoglycemia, congenital cardiovascular defects, congenital hypothyroidism, seizures, hypotonia, gastrointestinal problems, feeding difficulties, and increased susceptibility to infections.²² More than 75% of patients with KS harbor pathogenic variants in *KMT2D* (previously known as *MLL2*) located at 12q13.13 that typically occur de novo and in rare cases may be inherited (autosomal dominant Kabuki syndrome 1, KS1),^{23–26} and 5–8% have pathogenic variants in *KDM6A* located at Xp.11.3 that are most frequently de novo (X-linked dominant Kabuki syndrome 2, KS2).²⁷ Both male and female patients with *KDM6A*-associated KS have been reported; affected females may have skewed X-inactivation or have haploinsufficiency of the *KDM6A* protein.^{27,28} *KMT2D* and *KDM6A* encode for proteins that play an important role in the epigenetic regulation of transcriptionally active chromatin by interacting with each other in the protein complex known as ASCOM.²⁹ Pathogenic variants in these two genes cause abnormal chromatin remodeling, resulting in genome-wide effects impacting a range of organ systems. Fifteen to twenty percent of patients with a clinical diagnosis of KS may not carry variants in either *KMT2D* or *KDM6A* genes, respectively. In a more recent publication, other genes and loci that potentially contribute to a Kabuki-like syndrome have been discussed.²⁹

It was initially proposed that patients with KS caused by pathogenic variants in the *KMT2D* gene were more likely to be affected with classic facial dysmorphic features, in addition to a higher association with neonatal feeding problems, kidney abnormalities, skeletal joint dislocations, and palatal malformations.²⁵ A recent study hypothesized that

hypoglycemia due to HI may be a more common manifestation associated with KS2.³⁰ Although neonatal or infantile hypoglycemia seem to manifest in approximately 8–10% of patients with KS, the presentation of hypoglycemia secondary to HI in patients with KS was noted to be rare and found to have occurred in only 1 of 313 (0.3%) patients.³¹ In addition to hyperinsulinism, growth hormone deficiency and adrenal insufficiency have been proposed as possible contributing factors to persistent hypoglycemia in KS.^{25,30}

In this study, we characterized the presenting features of 10 newly identified individuals who came to medical attention due to neonatal hypoglycemia secondary to hyperinsulinism. Genetic evaluation revealed pathogenic variants in either *KDM6A* or *KMT2D* in all 10 cases, confirming a molecular diagnosis of KS. After the identification of these 10 cases, we adopted a retrospective approach and sought to determine the incidence of KS in HI patients who were diagnosed in the neonatal period. Thus, we evaluated 100 infants with a clinical diagnosis of HI and an unknown molecular diagnosis, for possible pathogenic variants in *KDM6A* and *KMT2D*.

PATIENTS AND METHODS

All patients involved in this study were diagnosed with persistent hyperinsulinism with onset in the neonatal period (Table 1). The patients were diagnosed with HI according to a set of criteria that included examination of the levels of plasma insulin, β -hydroxybutyrate, free fatty acids, and glycemic response to glucagon when plasma glucose was low.³² Of note, patients with HI do not necessarily present with high insulin levels, but they have an inappropriate level of insulin for their glucose measurements. Peripheral blood samples from patients 1–2 and 10 were referred to the molecular diagnostic laboratory at the University of Chicago for genetic evaluation of HI, and patient 3 was diagnosed with HI and referred for evaluation of KS at the University of Chicago. Patient 4 was identified at Boston Children's Hospital; patients 5–7 were identified at Cook Children's Medical Center and patients 8 and 9 were identified at the University of Exeter Medical School.

Of the 10 probands reported in this study, pathogenic variants in the KS genes, *KMT2D* and *KDM6A*, were identified by either array-comparative genomic hybridization (array-CGH) ($n = 2$, patients 1 and 2), Sanger sequencing of the KS genes ($n = 3$, patients 3, 8, and 9), targeted next-generation sequencing (NGS) of a HI gene panel ($n = 1$, patient 10), or clinical whole-exome sequencing (WES) ($n = 4$, patients 4–7). Patients 1 and 2 received array-CGH copy-number analysis of the associated HI genes (*ABCC8*, *KCNJ11*, *HADH*, *HNFI1A*, *HNFI4A*, *INSR*, *GCK*, *GLUD1*, *SLC16A1*, *UCP2*), and of the KS genes (*KDM6A*, *KMT2D*) using a high-resolution, custom-designed, exon-targeted 4 × 180 K array-CGH platform (Agilent Technologies, Santa Clara, CA). Patient 10 was tested on a targeted NGS panel that included 10 nonsyndromic HI genes (*ABCC8*, *KCNJ11*, *GLUD1*, *GCK*, *HADH*, *UCP2*, *INSR*, *HNFI1A*, *HNFI4A*, *SLC16A1*) and the KS genes (*KDM6A* and *KMT2D*).

Written informed consent was obtained to allow for the use of the medical history, genetic testing report, and photographs (if available) as approved by the institutional review board at the different institutions.

Retrospective analysis of HI infants for pathogenic variants in the KS genes

For the purpose of this study, we focused our retrospective analysis on patients who received negative testing results on a panel of 10 genes associated with nonsyndromic HI at the University of Chicago, and were less than 2 years old at the time of testing. Previous genetic analysis consisted of a combined targeted next-generation sequencing (NGS) panel³³ and copy-number analysis by array-CGH of 10 HI genes: *ABCC8*, *KCNJ11*, *HADH*, *HNF1A*, *HNF4A*, *INSR*, *GCK*, *GLUD1*, *SLC16A1*, and *UCP2*. We have recently updated this panel to include genes that are associated with syndromic forms of HI including the KS genes, *KDM6A* and *KMT2D*. From our patient database, we selected 100 patients that fit the above criteria and analyzed them on the updated HI panel to specifically identify mutations in the *KDM6A* and *KMT2D* genes.

RESULTS

Clinical details and genetic findings

Patient 1 presented with a biochemical profile consistent with HI and tachypnea on his first day of life (DOL1) (Table 1). He was able to achieve normoglycemia through diazoxide therapy. The patient was referred for testing of the original nonsyndromic HI gene panel by NGS and targeted array-CGH at 1 week of age, the results of which were negative. However, a hemizygous deletion of exons 3 through 24 of the *KDM6A* gene was identified by array-CGH due to the inclusion of these two genes on the array library design. The deletion was found to be in an apparent mosaic state (Fig. 1a). Duplex polymerase chain reaction (PCR) of exons 10 and 19 of the *KDM6A* gene was performed to confirm the presence of the *KDM6A* deletion in this patient. This analysis revealed the presence of a faint band corresponding to the *KDM6A* exons 10 and 19 in the patient's DNA (Fig. 1b). Sequence analysis performed on the PCR products obtained from the duplex PCR reactions using sequencing primers for *KDM6A* exons 10 and 19 confirmed the specificity of these products. These collective results provide confirmation of the *KDM6A* deletion in this patient and also suggest that the deletion likely occurred postfertilization and is thus present in a mosaic state in this individual. The detection of a pathogenic variant in *KDM6A* provided the patient with a molecular diagnosis of KS. His facial features were unremarkable during the neonatal period, however starting at age 4 months facial features become more characteristic for KS (Figure 1c and Table 2).

Patient 2 presented with persistent hypoglycemia due to HI on DOL1. The prenatal care was complicated by maternal alcohol exposure. She had an uncomplicated delivery, but was noted to have muscle hypotonia and labored breathing

following feedings. With diazoxide therapy she was able to achieve normoglycemia (Table 1). Similar to patient 1, genetic evaluation for HI was negative for 10 nonsyndromic HI genes analyzed by NGS and array-CGH, however a heterozygous deletion of exons 7–17 in *KDM6A* was detected through array-CGH (Fig. 1d). The patient was evaluated by an endocrinologist after the diagnosis of KS, noting dysmorphic features consisting of long eyelashes, low-set ears, and persistent finger pads (Table 2).

Patient 3 presented with hypoglycemia on DOL1 (Table 1) that was responsive to diazoxide treatment. At age 6 months, an evaluation by a geneticist noted additional findings including long palpebral fissures with lateral eversion of the lower eyelids, hirsutism, fifth finger clinodactyly, tapering fingers, and abnormal palmar creases (Fig. 1e and Table 2). In addition, renal hypoplasia, an additional renal cyst, and a sacral dimple were present in the patient. Chromosomal microarray analysis was normal. Sanger sequencing of *KMT2D* was performed due to clinical suspicion of KS and demonstrated a pathogenic two base pair duplication in *KMT2D*, c.603_604dup (p.Gly202Alafs*7).

Patient 4 presented with hypoglycemia on DOL1 and was treated adequately with diazoxide (Table 1). Her perinatal birth history was complicated by intrauterine growth retardation (IUGR) detected perinatally at 32 weeks gestation. Whole-exome sequencing (WES) performed at 1 year of age revealed a de novo heterozygous truncating variant in *KDM6A*, c.357C>G (p.Tyr119*). She manifested typical Kabuki facies (Fig. 1f), persistent finger pads, feeding difficulties, and short stature (Table 2).

Patient 5 experienced hypoglycemia shortly after birth on DOL1. He required intravenous fluids with a high glucose infusion rate of 20 mg/kg/min. He was diagnosed with HI on DOL 8, which was well controlled using diazoxide until 3 months of age (Table 1). WES performed at 1 year of age revealed a de novo truncating variant in *KMT2D*, c.11149C>T (p.Gln3717*). He manifested typical KS facies with long palpebral fissures, short nasal columella, arching eyebrows, in addition to developmental delay, feeding difficulty, hearing loss and atrial septal defect (Table 2).

Patient 6 had a perinatal history complicated by polyhydramnios and maternal history of asthma and tobacco use. She presented with hypoglycemia on DOL1 and was diagnosed with HI on DOL7. She was partially responsive to diazoxide and adequately managed with continuous overnight feeds. At 4 months of age there was evidence of diffuse disease so she received octreotide and achieved partial response. At 1 year of age she received a 98% pancreatectomy (Table 1). Genetic testing for the original nonsyndromic HI panel was negative. At 15–18 months of age she was evaluated by WES and found to harbor a truncating *KMT2D* variant, c.709del, (p.Glu237Serfs*24). She has typical KS facies with long palpebral fissures, short nasal columella, low-set ears, in addition to developmental delay, feeding difficulty, and hearing loss. At 2 years and 11 months of age, her hypoglycemia was managed medically with lantreotide,

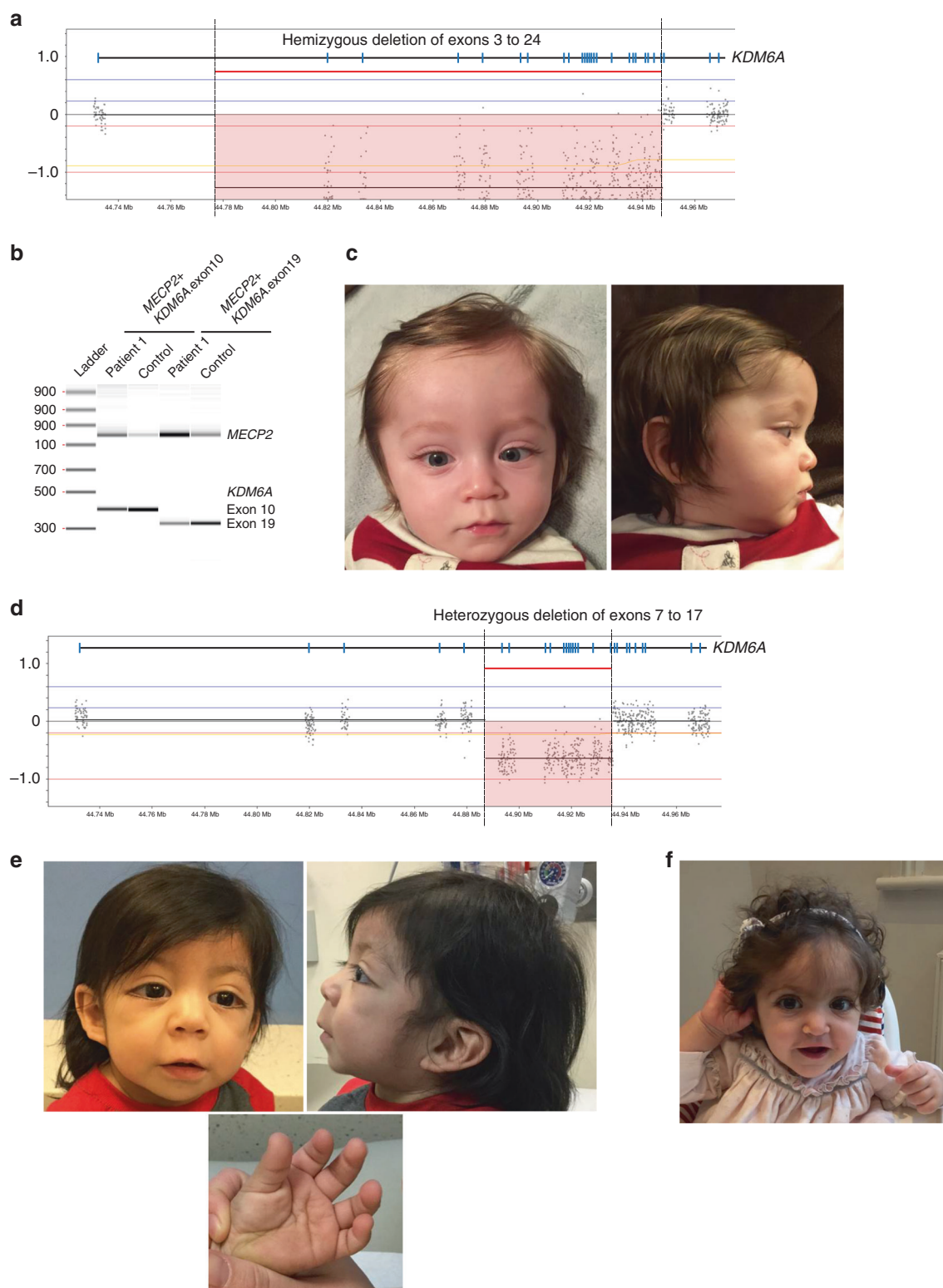


Fig. 1 Clinical features and/or molecular characterization of Patients 1–4. **a** Array-CGH results for patient 1 showing a hemizygous deletion involving exons 3–24 of *KDM6A* (arr[hg19] Xp11.3(44818602_44947539)x0). The reference sequence used for *KDM6A* is NM_021140.3. **b** Confirmation analysis of the deletion by duplex-PCR of exons 10 and 19 of *KDM6A* using a control amplicon in the *MECP2* gene revealed the presence of a faint band corresponding to the *KDM6A* exons 10 and 19 in the patient's DNA, suggesting the likely mosaic nature of the deletion. **c** Facial features of patient 1 at 9 months of age, showing long palpebral fissures, long eyelashes, and thin vermillion of the upper lip. **d** Array-CGH results for patient 2 providing evidence of a heterozygous deletion involving exons 7–17 of *KDM6A* (arr[hg19] Xp11.3(44892185_44935785)x1). **e** Patient 3 at 13 months old with the typical facial appearance and presence of persistent fingertip pads. **f** Patient 4 at 14 months old with the typical facial appearance of KS.

Table 1 Clinical history of 10 patients who presented with Congenital Hyperinsulinism (CHI)

	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5
Current Age	8 months	18 months	13 months	14 months	5 years
Gender	Male	Female	Male	Female	Male
Ethnicity & Family history	Italian/German/Slovakian, nonconsanguineous	Caucasian parents, maternal depression, history of alcohol abuse/dependence, gastric bypass, epilepsy, tobacco use 1/2 pack per day. Paternal heart condition, hypertrophic cardiomyopathy with defibrillator	Hispanic/Latino, maternal family history of asthma, paternal diabetes age of onset 28 years	Italian/Iranian, nonconsanguineous	Hispanic, nonconsanguineous
Age at delivery	37 5/7 weeks via NSVD to G1P0 28 years-old mother	38 6/7 weeks via NSVD to G5P0040 36 years-old mother	39 6/7 weeks via NSVD to G2P2 24-year-old mother	39 4/7 weeks via NSVD	39 weeks via NSVD
Birth weight	6 lbs 15 oz	7 lbs 3 oz	7 lbs 8 oz	6 lbs 0.7 oz	7 lbs 8 oz
Perinatal complications	Tachypnea requiring CPAP, hypoglycemia	Increased work of breathing after feeding	Desaturations on DOL3	IUGR at 32 weeks	—
Presentation of hypoglycemia	DOL1	DOL1	DOL1	DOL1	DOL8
Plasma glucose (mg/dL) Ref: 80–120[Lowest recorded]	51[29]	21[21]	44[12]	75.6[25.2/34.2]	47[40]
Insulin (uIU/ml = mU/L)	10.9	8.1	3.9	16.3	40
Beta hydroxybutyrate (mmol/L)	0.04	NA	0.15	NA	0.04
Free fatty acids (mmol/L)	0.28	NA	NA	NA	NA
Glycemic response to glucagon (mg/dL)	70	110 (taken during assessment for response to diazoxide)	Response to glucagon noted but no value available	NA	20
Treatment of hypoglycemia	Diazoxide 4.5 mg/kg/day. Started at 15 mg/kg/day	Diazoxide 5 mg/kg/day	Diazoxide 4 mg/kg/day	Initially with chlorothiazide 0.2 ml twice/day, diazoxide 3 mg/kg three times/day	Diazoxide until 3 months of age
Was normoglycemia achieved?	Yes	Yes	Yes	Yes	Yes

	Patient 6	Patient 7	Patient 8	Patient 9	Patient 10
Current Age	3 years	9 months	18 months	17 months	7 months
Gender	Female	Male	Female	Female	Male
Ethnicity & Family history	Caucasian	Asian	Caucasian, nonconsanguineous	Iranian/Italian, nonconsanguineous	Ecuadorian
Age at delivery	39 weeks via NSVD	37 weeks via C-section due to nonreassuring fetal heart tones	39 1/7 weeks via elective C-section for breech presentation	39 4/7 weeks	38 weeks via C-section
Birth weight	7 lbs 14 oz	5 lbs 5 oz	11 lbs 2 oz	6 lbs 4 oz	6 lbs 8 oz
Perinatal complications	Polyhydramnios with tobacco use and asthma in the mother	Single umbilical artery, IUGR, and nuchal cord	PPHN requiring sildenafil	None	15-day NICU stay for respiratory distress colostomy at DOL 1, Surgeries: colostomy, PSARP, g-tube and fundoplication
Presentation of hypoglycemia	DOL7	DOL7	DOL1	DOL1	Likely early DOL but only discovered while coming from Ecuador for ostomy reversal during preoperative work at 7 months old
Plasma glucose (mg/dL) Ref:	35[31]	46[30]	17	25	41[33]

Table 1 continued

	Patient 6	Patient 7	Patient 8	Patient 9	Patient 10
80–120[Lowest recorded]					
Insulin (uIU/ml = mU/L)	8.8	1	8.5	16.3	6.9
Beta hydroxybutyrate (mmol/L)	0.21	0.22	0.6	0.4	0.6
Free fatty acids (mmol/L)	1.82	NA	0.3	0.6	1.3
Glycemic response to glucagon (mg/dL)	42	82	NA	NA	80
Treatment of hypoglycemia	Diazoxide (partially responsive), octreotide, pancreatectomy due to diffuse disease, at 3 years old managed with Somatuline, g-button feeds, overnight feeds	Diazoxide, responsive and well controlled. At 4 months of age stopped due to pulmonary hypertension. Solcarb to help maintain blood glucose. Now managed with feeds	Partial pancreatectomy and diazoxide 10 mg/kg/day	Diazoxide 3 mg/kg/day	Diazoxide 10 mg/kg/dose three times a day
Was normoglycemia achieved?	No	Yes	Yes	Yes	Yes

Laboratory values for plasma glucose, insulin, beta-hydroxybutyrate, and free fatty acids were recorded during a critical sample collection
NSVD normal spontaneous vaginal delivery, *CPAP* continuous positive airway pressure, *IUGR* intrauterine growth retardation, *DOL* day of life, *PPHN* persistent newborn pulmonary hypertension, *PSARP* posterior sagittal anorectoplasty, *NICU* neonatal intensive care unit, *CHI* congenital hyperinsulinism

g-button feeds, and overnight feeds. She continued to have recurrent episodes of hypoglycemia (Tables 1 and 2).

Patient 7 had a perinatal history complicated by a single umbilical artery, IUGR and nuchal cord. During the neonatal period he was diagnosed with HI at DOL7, and was responsive to diazoxide therapy. Genetic evaluation for the original nonsyndromic HI panel was negative. At 4 months of age he developed pulmonary hypertension, which was resolved by discontinuing diazoxide therapy. His blood glucose was then maintained by Solcarb and at 9 months of age his glucose levels were well managed by feeds (Table 1). WES was performed at 9 months of age and identified a de novo *KMT2D* variant, c.8366G>A (p.Arg2789Gln). This sequence change alters the last nucleotide of exon 33 of *KMT2D*. In silico splicing prediction algorithms (MaxEntScan, Human Splicing Finder, and NNSplice) indicate that this sequence change is predicted to weaken the canonical acceptor splice site, and it is thus likely to result in aberrant splicing of *KMT2D* mRNA. The patient at 9 months of age was noted to have typical KS facies with long palpebral fissures, in addition to developmental delay, feeding difficulty, and a sacral dimple (Table 2).

Patient 8 had a birth complicated by persistent pulmonary hypertension of the newborn (PPHN), which required the use of sildenafil. During the neonatal period she had feeding difficulties and presented with HI at DOL1. She received a

partial pancreatectomy and her condition was well controlled with the use of diazoxide (Table 1). Genetic evaluation for the original nonsyndromic HI panel at University of Exeter (*KCNJ11*, *ABCC8*, *GLUD1*, *HADH*, *GCK*, *HNF4A*, *INSR*, *SLC16A1*, *TRMT10A*, and *HNF1A*) was negative. At 11 months of age due to the observation of Kabuki-like features, she received genetic testing for KS that revealed a de novo truncating variant in *KDM6A*, c.2074_2075del (p.Gln692Glyfs*37). She was noted to have dysmorphic facial features that include a depressed nasal bridge, prominent eyes, and a small jaw, and exhibited premature thelarche. She also had skeletal abnormalities, specifically a left developmental dislocation of the hip (DDH) that was treated with a Pavlik harness, in addition to a sacral dimple. Her cardiovascular abnormalities include a patent ductus arteriosus (PDA), and a small patent foramen ovale (PFO) with mild concentric left ventricular hypertrophy (LVH) of the heart (Table 2).

Patient 9 was born with no known perinatal complications. She presented with hypoglycemia due to HI at DOL1 that was well controlled with diazoxide therapy (Table 1). At 7 months of age she was noted to have dysmorphic features consistent with KS. Genetic testing for *KDM6A* and *KMT2D* identified a de novo truncating variant in *KDM6A*, c.357C>G (p.Tyr119*). She was noted to have long eyelashes, high arch palate and a small jaw, with reduced subcutaneous fat on her hands and truncal hypotonia with mild developmental delay.

She also exhibited feeding difficulties with poor weight gain and is of short stature (Table 2).

Patient 10 was born with an imperforate anus, undescended testes, and affected with congenital torticollis and a ventricular septal defect (VSD). He was hospitalized for 15 days after birth for respiratory distress and received a colostomy procedure at DOL1. At 7 months of age he was scheduled to receive an ostomy reversal procedure, and was found during preoperative laboratory work-ups to have severe hypoglycemia. He was responsive to diazoxide treatment and endocrinological evaluation indicated a diagnosis of HI (Table 1). Genetic evaluation for 12 HI genes including the 2 additional KS genes identified a heterozygous pathogenic *KMT2D* variant, c.6613delinsAA (p.Ala2205Asnfs*38). He was noted to have high arched eyebrows, long eyelashes, and blue sclerae. He also exhibited developmental delay, and is of low weight and height (Table 2).

Retrospective analysis of HI infants for pathogenic variants in the KS genes

To determine the frequency of KS in patients who presented with HI, we reevaluated 100 patients who received a diagnosis of HI under 2 years of age and also tested negative in our laboratory using the original 10-gene nonsyndromic HI panel, which did not include the KS genes at the time of testing. We identified one patient out of a hundred, who harbored a pathogenic variant in *KMT2D*, c.5135del (p.Lys1712Argfs*10), leading to a molecular diagnosis of KS in this patient (95% confidence interval [CI]: 0.0018–0.0545). This child presented at birth with severe and persistent hypoglycemia and was diagnosed with HI shortly after birth. He was responsive to diazoxide and at age 3 has continued successfully on this treatment. He has classical clinical features of KS including long eyelashes, epicanthus, long palpebral fissures with upslant, large slightly low set ears, and fingertip pads. In addition to the HI, he has developmental delays and feeding difficulties.

DISCUSSION

Genetic defects in genes regulating insulin secretion are responsible for monogenic, non-syndromic forms of persistent HI, although metabolic conditions in the newborn or birth complications in a neonate can also cause presentation of HI.¹⁶ In addition, a variety of multisystemic syndromes have also been associated with HI. Kabuki syndrome is associated with the presentation of HI (HI incidence 1:50,000),¹⁵ but overall it is a rare condition, with an estimated incidence of 1:32,000 in the Japanese population and 1:86,000 in Australia and New Zealand.^{20,34} A typical findings have been documented and persistent hypoglycemia due to HI has been described as an uncommon occurrence in KS that has been noted in a limited number of patients, although transient neonatal or infantile hypoglycemia had been observed in as much as 8–10% of patients with KS.³¹ In addition, hypoglycemia in patients with KS has also been attributed to combined pituitary hormone deficiency,²⁴

growth hormone deficiency,²⁵ and adrenal insufficiency, respectively.^{35,36}

In this study we describe 10 unrelated patients with molecularly confirmed KS who presented with persistent HI during the neonatal period. To assess the incidence of KS in a cohort of unselected HI patients, we retrospectively analyzed 100 individuals of less than 2 years of age who received negative tests results on a HI NGS panel of 10 nonsyndromic genes, for the presence of pathogenic variants in *KMT2D* and *KDM6A*. This analysis identified an additional patient who harbored a pathogenic variant in *KMT2D*, leading to a diagnosis of KS. Of the 11 pathogenic variants identified, 5 were truncating variants in *KMT2D*, 1 was a splice variant in *KMT2D*, 3 were truncating variants in *KDM6A*, and 2 were intragenic deletions in *KDM6A*, with 1 being a mosaic deletion. To the best of our knowledge, all variants with the exception of c.11149C>T (p.Gln3717*)²⁴ are novel to this study. Our results strongly suggests that genetic evaluation for KS genes, including sequencing and deletion duplication analysis should be carried out during testing for genetic etiologies of HI in neonates, as this approach may inadvertently uncover early diagnoses of KS.

The majority of patients with KS harbor pathogenic variants in *KMT2D* and only 2–8% of cases are due to pathogenic variants in the *KDM6A* gene.³⁷ In our cohort ($n = 11$), we observed 5 pathogenic variants (45.5%) in *KDM6A*, which is evident enrichment over the general frequency of *KDM6A* pathogenic variants (2–8%) in KS, suggesting that patients with KS harboring pathogenic *KDM6A* variants (KS2) may have an increased likelihood of presenting with HI as compared with patients with KS who harbor pathogenic variants in *KMT2D* (KS1). This is an intriguing finding that warrants further investigations in a larger cohort of KS patients, but emphasizes the need for clinicians to more carefully evaluate KS2 patients for hypoglycemic episodes.

Overall, persistent hypoglycemia secondary to HI has been described as an atypical finding in patients with KS.³¹ In a study by Genevieve et al., the frequency of neonatal hypoglycemia among patients with KS was 21/313 (6.7%); however, HI was suspected in only one patient (0.3%), although it is unclear if a proper assessment of HI was conducted for each of these patients in this study.³¹ The presentation of HI in patients with KS has also only been documented in single case reports.^{4,38,39} Our data indicate that the rate of HI among patients with KS may be higher than previously suggested as more patients undergo genetic evaluation. It is possible that only the most severe and persistent HI cases may be documented, as milder or transient forms of HI cases may remain undetected. In our retrospective study on the cohort of 100 HI patients with negative molecular testing at our institution, we identified a single patient with a molecular diagnosis of KS. This indicates that the incidence of KS in neonates with HI without an identifiable genetic cause may be around 1% (95% CI: 0.0018–0.0545), although the number of studied cases is too small to draw any final conclusions.

Table 2 Kabuki syndrome features of the 10 patients

Features	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5
Age of KS diagnosis	2 months	18 months	6 months	6 months	1 year
Genotype	Mosaic hemizygous deletion of <i>KDM6A</i> exons 3–24 arr[hg19]Xp11.3(44818602_44947539)x0	De novo deletion of <i>KDM6A</i> exons 7–17 arr[hg19] Xp11.3 c.603_604dup (p.(44892185_44935785)x1 Gly202Alafs*7)	De novo <i>KMT2D</i> c.603_604dup (p. Gly202Alafs*7)	De novo <i>KDM6A</i> c.357C>G (p.Tyr119*)	De novo <i>KMT2D</i> c.11149C>T (p. Gln3717*)
Facial	Long palpebral fissures	Long eyelashes, low-set ears, protruding ear lobes	Long palpebral fissures with lateral eversion of eyelid	Long palpebral fissures, eversion of lateral third of lower eyelid, arched/sparse eyebrows, depressed/flat nasal tip, large/dysmorphic ears	Long palpebral fissures, short nasal septum, arching eyebrows
Eye	Left homonymous hemianopsia visual field defect, exotropia (since resolved), and bilateral hyperopia	Left-side strabismus	–	–	–
Oral	Thin upper lip vermillion	–	Thin upper lip vermillion	–	–
Skeletal	Hip dysplasia	–	Sacral dimple with hair tuft	Hip dysplasia	–
Hand	–	Fleshy finger pads	5th finger clinodactyly, tapering fingers, abnormal palmar creases, fleshy finger pads	Fetal fingertip pads	–
Neurological	Congenital partial agenesis of the corpus callosum with right paramedian posterior interhemispheric cyst, developmental delay, seizures	Decreased symmetric muscle tone and strength	Developmental delay	Neonatal hypotonia	Hearing loss, developmental delay
Cardiovascular	Small atrial septal defect and resolving biventricular hypertrophy	–	–	–	Atrial septal defect
Gastrointestinal	Feeding difficulties and gastroesophageal reflux	–	Feeding difficulties	Feeding difficulties and gastroesophageal reflux	Feeding difficulties
Urogenital	–	–	Dysplastic left kidney, renal cysts in right	–	–
Endocrine	Short stature: height <5th percentile	–	Short stature: 5th percentile in height	Short stature	–

Features	Patient 6	Patient 7	Patient 8	Patient 9	Patient 10
Age of KS diagnosis	15–18 months	9 months	11 months	7 months	7 months
Genotype	De novo <i>KMT2D</i> c.709del (p. Glu237Serfs*24)	De novo <i>KMT2D</i> c.8366G>A (p.Arg2789Gln)	De novo <i>KDM6A</i> c.2074_2075del (p. Gln692Glyfs*37)	De novo <i>KDM6A</i> c.357C>G, (p.Tyr119*)	De novo <i>KMT2D</i> c.6613delinsAA, (p. Ala2205Asnfs*38)
Facial	Long palpebral fissures, short nasal septum, low set ear	Long palpebral fissures	Depressed nasal bridge	Long eyelashes	Arched eyebrows
Eye	–	–	Prominent eyes	–	Long eyelashes, blue sclerae
Oral	–	–	Small jaw	High arch palate, small jaw	–

Table 2 continued

Features	Patient 6	Patient 7	Patient 8	Patient 9	Patient 10
Skeletal	-	Sacral dimple	Left DDH with Pavlik harness	-	-
Hand	-	-	-	Reduced subcutaneous fat	-
Neurological	Hearing loss, developmental delay	Developmental delay	Sacral dimple	Truncal hypotonia, mild developmental delay	Developmentally delayed
Cardiovascular	-	One umbilical artery	PDA and small PFO with mild concentric LVH	-	Heart murmur, ventricular septal defect
Gastrointestinal	Feeding difficulties	Feeding difficulties	Feeding difficulties, cows milk protein intolerance	Feeding difficulties	Imperforate anus, GERD
Urogenital	-	-	-	-	Undescended testes
Endocrine	-	-	Premature thelarche	Poor weight gain and short stature	Low weight and height

DDH developmental dysplasia of the hip, PDA patent ductus arteriosus, PFO patent foramen ovale, GERD gastroesophageal reflux disease
The reference sequence used for *KDM6A* is NM_021140.3. The reference sequence used for *KMT2D* is NM_003482.3

The 10 patients on whom clinical information was available were documented to have dysmorphic features typical of KS and exhibited developmental delays and feeding difficulties. This is in agreement with previous reports documenting typical phenotypic findings in patients with KS and HI. Patient 1 received a relatively early diagnosis of KS as he was undergoing genetic evaluation for HI. The other patients were diagnosed in the range of 6 months to 18 months in age. This agrees with previous observations that the early diagnosis of KS in the neonatal period is challenging as the typical facial features of KS may not be not clearly evident in young children.³⁷ For a patient with an early diagnosis of KS (especially for KS2), it is important to monitor for possible hypoglycemic episodes that could aggravate their neurological development. Conversely, for a neonate who presents with HI, it is important to consider KS as one of the differential diagnoses. An early diagnosis of KS in the neonatal period can prompt further clinical evaluation for other underlying conditions that may be present in patients with KS, such as congenital heart defects and hip dysplasias that require prompt medical attention. Early diagnosis of KS after an initial presentation of HI also allows the family to receive appropriate prognostic counseling, and could potentially prompt earlier referral to special education or early intervention services to allow maximum developmental achievement for these patients, who are at risk for delays and intellectual disability.

Although the etiology of HI in patients with KS is still unclear, we observe that in the majority of our patients (9/10), the HI was adequately managed with the use of diazoxide, as similarly described in a previous publication.⁴ Because hypoglycemia in HI of patients with KS can be managed using diazoxide, timely diagnosis of the condition is crucial to improving outcomes for these patients.

In conclusion, the results of this study indicate that patients with KS caused by pathogenic variants in *KDM6A* (KS2) may be at higher risk to develop HI during the neonatal period, than those with pathogenic variants in *KMT2D* (KS1). Patients with KS with a history of HI do not appear to have additional specific phenotypic features that would differentiate them from patients without HI. In patients with a diagnosis of KS, awareness of the risk of HI and early recognition of hypoglycemic episodes is important to mitigate the potential neurologically damaging effects of untreated hypoglycemia. The typical features of KS, including the characteristic dysmorphic facial features, may be challenging to recognize in the neonatal period. Thus, we recommend considering KS in the differential diagnosis of persistent HI and also including the comprehensive evaluation (sequencing and deletion/duplication analysis) of the KS-associated genes *KMT2D* and *KDM6A* during genetic testing for HI. Early diagnosis may aid in appropriate follow-up and treatment of the hypoglycemic events in patients with KS, since these patients are likely to respond to the use of diazoxide. With increased genetic testing for KS in patients with HI, we may find that the presentation of HI in patients with KS, though an infrequent occurrence, could be more common than previously postulated.

ACKNOWLEDGEMENTS

S.E. is a Wellcome Trust Senior Investigator. We would like to acknowledge the valuable participation of the patients and their families in our study.

DISCLOSURE

The authors declare no conflicts of interest.

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