CLINICAL REPORT

AMERICAN JOURNAL OF medical genetics PART A

Hypoglycemia in Kabuki Syndrome

Anbezhil Subbarayan[1] and Khalid Hussain[1,2]*

[1]Department of Pediatric Endocrinology, Great Ormond Street Hospital, London, UK

[2]Developmental Endocrinology Research Group, Molecular Genetics Unit, Institute of Child Health, University College London, London, UK

Manuscript Received: 18 January 2013; Manuscript Accepted: 8 August 2013

Kabuki syndrome (KS) is a congenital malformation disorder with a spectrum of clinical manifestations involving different organs. Until the identification of *MLL2* gene mutation in 2010, the diagnosis was made only clinically by the characteristic facial features with other common and uncommon features. Hypoglycemia, although an uncommon feature in KS, is very important to be recognized, as early diagnosis and appropriate management will reduce further long-term neurologic morbidity in these patients. We report on four patients with KS presenting with persistent hypoglycemia. Hyperinsulinemic hypoglycemia was the cause of hypoglycemia in two out of four patients and one patient had growth hormone deficiency. The mechanism of the hypoglycemia in one patient is still unclear. Three out of these four patients were found to have mutation in the *MLL2* gene. Our observations suggest that patients with KS may have hypoglycemia due to different mechanisms and that *MLL2* gene may have a role in glucose physiology. © 2013 Wiley Periodicals, Inc.

Key words: Kabuki syndrome; hypoglycemia; hyperinsulinemic hypoglycemia; growth hormone deficiency

How to Cite this Article:
Subbarayan A, Hussain K. 2014.
Hypoglycemia in Kabuki syndrome.
Am J Med Genet Part A 164A:467–471.

INTRODUCTION

Kabuki syndrome (KS, OMIM #147920) is a congenital malformation disorder with a wide spectrum of clinical manifestations involving multiple organ systems. KS was first described simultaneously by two authors from Japan in 1981 [Kuroki et al., 1981; Niikawa et al., 1981]. The estimated prevalence of KS is about 1:32,000 [Niikawa et al., 1988] but now around 400 cases has been described in the literature across the world involving many ethnic groups [Matsumoto and Niikawa, 2003; Bokinni, 2012].

KS is characterized by typical facial features with long palpebral fissures, eversion of lateral one-third of lower eyelid, arched eyebrows, short columella with depressed nasal tip and prominent ears. This is associated with other common features like skeletal anomalies, dermatoglyphics abnormalities, short stature, learning difficulties along with some uncommon features as summarized in Table I [Adam and Hudgins, 2004]. Six to eight percent of patients with KS present with hypoglycemia either in the neonatal period or later in their life [Genevieve et al., 2004; Armstrong et al., 2005].

Hypoglycemia can be transient or persistent in KS but its etiology and management have not been fully described. We report on four interesting cases of KS with persistent hypoglycemia with varied etiology, their management and response to treatment. Early recognition and prompt treatment of hypoglycemia is very important to prevent further long-term brain damage in patients with KS who are already compromised with high prevalence of learning difficulties. The major breakthrough in the etiology of KS occurred when mutations in *MLL2* gene were identified in 2010 [Ng et al., 2010]. All our patients have been tested for mutations in the *MLL2* gene. Another remarkable recent development in the etiology of KS is the identification of mutation in the X chromosome gene *KDM6A* in few patients in whom mutations in the *MLL2* gene were not found [Lederer et al., 2012].

CLINICAL REPORT

We report on four patients who were clinically diagnosed with KS and their clinical features are features summarized in Table I. Each of these patients presented with hypoglycemia on day 1 of life and the biochemical findings at the time of hypoglycemia are shown in Table II.

Patient 1

This 8-year-old boy, who was diagnosed with KS at the age of 1 year, presented with hypoglycemic seizures on day 1 of life. He was born by normal delivery at 39 weeks gestation with birth weight of 3.1 kg (−0.7 SD) with no immediate perinatal complications. His hypoglycemic screen showed inappropriately raised serum insulin levels with suppressed lipolysis and ketogenesis (Table II). Given these biochemical features he was diagnosed with hyperinsulinemic

*Correspondence to:
Dr. K. Hussain, Developmental Endocrinology Research Group, Molecular Genetics Unit, Institute of Child Health, University College London, 30 Guilford Street, London WC1N 1EH, UK.
E-mail: khalid.hussain@ucl.ac.uk
Article first published online in Wiley Online Library (wileyonlinelibrary.com): 5 December 2013
DOI 10.1002/ajmg.a.36256

TABLE I. Clinical Features

Features	Patient 1	Patient 2	Patient 3	Patient 4
Age (years)	8.6	3	7	1.9
Gender	Male	Male	Female	Male
Characteristic face				
Long palpebral fissures	+	+	+	+
Eversion of lateral third of lower eyelid	−	+	+	+
Arched/sparse eyebrows	+	+	+	+
Depressed/flat nasal tip	+	+	+	+
Large ears/dysmorphic ears	−	−	−	+
Others	Microcephaly	Trigonocephaly	−	−
Eye abnormalities				
Eye anomaly	−	−	−	+
Visual impairment	+	−	−	+
Others	Rod dystrophy	−	−	Cataract
Otologic findings				
Otitis media	−	−	−	+
Hearing loss	+	−	−	+
Oral abnormalities				
Cleft palate	−	−	+	−
Abnormal dentition	+	−	−	−
Thin upper lip vermilion	−	−	−	+
Lip nodules	−	−	−	−
Skeletal abnormalities				
Scoliosis	+	−	−	−
Hip dysplasia	−	−	−	−
Joint hypermobility	+	+	+	+
Others	−	−	−	Vertebral abnormalities
Hands				
Brachydactyly	−	+	−	−
Fetal fingertip pads	+	−	−	+
Neurological problems				
Developmental delay	+	+	+	+
Learning difficulties	+	+	+	+
Seizures	−	−	+	−
Neonatal hypotonia	+	−	−	+
Cardiovascular				
Structural abnormalities	−	−	+	+
Gastrointestinal problems				
Feeding difficulties	+	+	+	+
Gastroesophageal reflux	+	+	+	+
Others	−	−	−	Enteropathy
Urogenital				
Kidney malformation	−	+	+	−
Endocrine problems				
Hypothyroidism	−	−	+	−
Short stature	−	−	+	+
Others	−	−	−	Growth hormone deficiency
Immunological problems				
Frequent infections	+	−	+	+
Others	IgA deficiency	−	IgA deficiency	Hypogamma globulinemia
Additional features	Spigelian hernia	Diaphragmatic hernia	SLE hypertension	Multiple food allergy

+, Present; −, not present.

hypoglycemia and was commenced on therapy with Diazoxide and Chlorothiazide from the age of 3 weeks. The dose of Diazoxide was titrated to a maximum of 10 mg/kg/day to maintain normal blood glucose levels. He continued on Diazoxide for 4 years and was then gradually weaned off over a period of few months and stopped at the age of five years. After stopping his medications his 24-hr blood glucose profile was normal (all blood glucose levels >3.5 mmol/L) and he managed to fast for 18 hr with no evidence of hypoglycemia.

TABLE II. Features of Hypoglycemia in KS

Features	Patient 1	Patient 2	Patient 3	Patient 4
Age of onset	Day 1	Day 1	Day 1	Day 1
Presenting symptoms	Seizure	Jitteriness	Poor feeding	Poor feeding
Investigations				
Blood glucose (mmol/L)	2.4	2.5	2.3	2.9
Insulin (mU/L)	22	3.9	42.3	<2
NEFA (mmol/L)	0.32	0.38	1.67	1.3
3-BOHB (mmol/L)	0.08	0.07	1.08	1.02
Cortisol (nmol/L)	454	317	610	221
Growth Hormone (μg/L)	—	10.8	5.2	2.9
Lactate (mmol/L)	—	2.2	1.6	1.2
Ammonia (mcg/dl)	—	40	35	37
Cause of hypoglycemia	Hyperinsulinism	Hyperinsulinism	Not known	Growth hormone deficiency
Treatment	Diazoxide + Chlorothiazide	Diazoxide + Chlorothiazide	Feeding regimen	Growth hormone
MLL2 mutation	Negative	Positive, c.6971dupC heterozygote	Positive, c.5845C>T heterozygote	Positive, c.2992C>G and c.12964C>T heterozygote

NEFA, nonesterified fatty acids; 3-BOHB, 3-beta hydroxybutyrate.

His serum insulin levels were undetectable at the end of the fast and he showed an appropriate increase in the blood ketone levels suggesting that his hyperinsulinemic hypoglycemia had resolved. He continued to maintain normoglycemia since then. He was found to be negative for *ABCC8* and *KCNJ11* gene mutations known to cause congenital hyperinsulinism. No mutations in the *MLL2* gene have been found yet.

Patient 2

This 3-year-old boy was diagnosed with KS at the age of 1 year and later found be positive for *MLL2* mutation. He was born by normal delivery at 38 weeks gestation with birth weight of 2.9 kg (−0.71 SDS) with no perinatal complications. He presented on day 1 of life with jitteriness and poor feeding. His blood glucose level was low and the hypoglycemia screen showed inappropriately raised serum insulin levels (Table II). He was diagnosed with hyperinsulinemic hypoglycemia and was commenced on Diazoxide and Chlorothiazide. His dose of Diazoxide was gradually increased up to a maximum of 7 mg/kg/day to normalize his blood glucose. He continues to maintain normoglycemia on the above dose of Diazoxide. No mutations in *ABCC8* and *KCNJ11* gene detected.

Patient 3

This 7-year-old girl was born at 37 weeks gestation by normal delivery with a birth weight of 2.82 kg (−0.14 SDS). She was diagnosed with KS at the age of 18 months and later noted to have a mutation in the *MLL2* gene. She presented to her local hospital with poor feeding on day 1 and was noted to be hypoglycemic. Her blood glucose level normalized after increasing her feed volume and no further investigations was carried out at that time. She re-presented to our hospital at the age of 5 years with hypoglycemia. On further investigations, her blood glucose level dropped to 2.4 mmol/L at 14 hr of controlled fast with raised insulin levels suggestive of hyperinsulinemic hypoglycemia. However interestingly she also showed an appropriate increase in her blood ketones and fatty acids which was not typical of hyperinsulinism. Her metabolic investigations including lactate, ammonia, acyl carnitine, plasma amino acids, urine amino, and organic acids were normal (Table II). Her blood glucose levels are maintained at normal levels by giving her continuous feeds at night and regular bolus feeds during the day through gastrostomy. The mechanism of the hypoglycemia is still unclear in this patient.

Patient 4

This is a 22-month-old boy, first born to nonconsanguineous Caucasian parents. He was born by ventouse delivery at 38 weeks gestation with birth weight of 3.52 kg (0.7 SDS). He presented with hypoglycemia on day 1 of life which was managed at the local hospital with a regular feeding regimen. The family were then lost to follow up but then represented at the age of 6 months with intermittent hypoglycemia. On further investigations, he managed to fast only for 3 hr when his blood glucose dropped to 2.9 mmol/L with undetectable insulin levels and raised blood ketones and fatty acids. Interestingly his growth hormone and cortisol levels were low at the time of hypoglycemia (Table II). He was diagnosed with KS at the age of 8 months and was later found to be positive for *MLL2* mutation. He was also noted to have low serum IGF-1 (34 ng/ml) level and his growth velocity was low. He underwent a Glucagon stimulation test and was diagnosed with growth hormone deficiency with the growth hormone peak of 5 mcg/L (Normal: GH peak $\geq$ 6.7 mcg/L or 20 mU/L) and cortisol peak of 494 nmol/L (Normal: cortisol peak $\geq$ 450 nmol/L). His glucose profile improved significantly on growth hormone treatment and he was able to fast for 7 hr with no hypoglycemia at the age of 20 months.

METHODS

Sequencing of the Protein-Coding Part of *MLL2*

All coding 54 exons and splice junctions of *MLL2* were amplified by polymerase chain reaction and subsequently sequenced by capillary (Sanger) sequencing. The identified mutations were tested for a de novo origin when parental DNA was available. For missense mutations pathogenicity was tested using silico prediction tools (SIFT and Mutation Taster).

Results for Genetic Sequencing of *MLL2* Gene

In Patient 1, no mutations were identified in the coding regions of the *MLL2* gene. Sequencing analysis of the *MLL2* gene in Patient 2 showed a heterozygous change (c.6971dupC) which causes a frameshift change and is predicted to lead to premature termination of the translation (p.Asp2325X). This truncating mutation was considered to be highly pathogenic. In Patient 3, a heterozygote mutation (c.5845C > T) in the *MLL2* gene was found. This nonsense mutation was predicted to cause glutamine to be substituted for a stop codon at residue 1949 (p.Gln1949X). This truncating mutation was considered to be highly likely to be pathogenic. Finally in Patient 4, sequencing analysis of the *MLL2* gene showed a heterozygous c.12964C > T nonsense mutation that was predicated to substitute glutamine with a stop codon at residue 4322 (p.Gln4322X). This truncating mutation was considered to be highly pathogenic. This patient was also found to be heterozygous for the c.2992C > G missense change that was predicated to substitute proline with alanine a codon 998 (p.Pro998Ala). This variant has not been previously reported and in silico prediction software provided conflicting information on its likely pathogenicity.

DISCUSSION

Patients with KS can present with hypoglycemia which can be transient or persistent [Genevieve et al., 2004; Armstrong et al., 2005]. All of our four patients presented with hypoglycemia on day 1 of life. Patients 1 and 2 were diagnosed with hyperinsulinemic hypoglycemia within first few weeks of life and started treatment. So far there are only two reported cases of hyperinsulinemic hypoglycemia in KS [Ming et al., 2001a; Armstrong et al., 2005] but the management was not clearly explained and they were not tested for *MLL2* mutation. Hyperinsulinemic hypoglycemia is one of the commonest causes of persistent hypoglycemia in patients with KS and hence a high degree of suspicion is needed for early diagnosis and appropriate management. This is especially important as delay in the diagnosis and management of hyperinsulinemic hypoglycemia is associated with brain damage.

Interestingly, both our patients with hyperinsulinemic hypoglycemia responded to therapy with Diazoxide and Chlorothiazide and one of them needed medications only until 5 years of age. Patient 3 had intermittent hypoglycemia with no clear etiology identified and was managed by feeding regimen. Similar cases have been reported in the literature [Genevieve et al., 2004]. This patient has been extensively investigated so far but no underlying cause of the hypoglycemia has been found. Given that this patient is positive for the *MLL2* gene mutation she may well have a new mechanism of the hypoglycemia.

In Patient 4, hypoglycemia was secondary to growth hormone deficiency which was managed with growth hormone replacement therapy. Few cases of KS with Growth hormone deficiency have been reported in the literature so far [Niikawa et al., 1988; Gabrielli et al., 2002; Armstrong et al., 2005; Schrandr-Stumpel et al., 2005]. There was one patient with KS previously reported with adrenocorticotropic hormone (ACTH) deficiency as the cause of persistent hypoglycemia [Ma et al., 2005]. It is fascinating to find that even in such a rare condition the cause of hypoglycemia is diverse and hence complete evaluation of the patients are needed for appropriate management.

Following the identification of *MLL2* gene mutation in 2010 in patients with KS [Ng et al., 2010] this has been identified in 66–75.6% of patients with KS [Hannibal et al., 2011; Paulussen et al., 2011; Banka et al., 2012]. *MLL2* gene was first described in 1999 and encodes a large protein that is part of SET family of proteins [FitzGerald and Diaz, 1999]. The *MLL2*-knockout mice models showed there were 20 genes significantly down-regulated which comprised various functions including cell migration, growth adhesion, and transcriptional regulation [Issaeva et al., 2007]. *MLL2* is a transcriptional activator that induces the transcription of target genes by covalent histone modification and appears to be involved in the regulation of adhesion-related cytoskeletal events, which might affect cell growth and survival [Issaeva et al., 2007]. How this transcriptional activator activity of *MLL2* might impact pancreatic insulin release and cause hyperinsulinemic hypoglycemia is still unclear.

Most recently deletion of X chromosome gene *KDM6A* was identified in three patients with KS [Lederer et al., 2012] and point mutations in the same gene has been found in three more patients [Miyake et al., 2013]. *KDM6A* along with *MLL2* gene plays a role in embryogenesis and development. As this only a very recent development, our patients were not tested for the *KDM6A* gene mutation. In future this should be taken into consideration and would be interesting to explore any phenotype–genotype correlation for any mutation found in the *KDM6A* gene in patients presenting with hypoglycemia.

In our case report, three out of four patients who presented with hypoglycemia are found to be positive for the *MLL2* mutation. Even in the three patients with mutation positive, the mutations identified were different and the causes of hypoglycemia were also different making it difficult to explain a single molecular process in the pathogenesis. Further studies are required to understand how genetic defects in *MLL2* and *KDM6A* lead to defects in the physiology of glucose homeostasis.

In conclusion, hypoglycemia should be suspected in all patients with KS presenting with seizures and other nonspecific symptoms. Hyperinsulinemic hypoglycemia is an important cause of hypoglycemia in patients with KS and they respond well to medical management. Early diagnosis and appropriate management will benefit these patients by reducing further neurological damage.

REFERENCES

Adam MP, Hudgins L. 2004. Kabuki syndrome: A review. Clin Genet 67:209–219.

Armstrong L, Moneim A, Aleck K, Aughton D, Baumann C. 2005. Further delineation of Kabuki syndrome in 48 well-defined new individuals. Am J Med Genet Part A 132A:265–272.

Banka S, Veeramachaneni R, Reardon W, Howard E, Bunstone S, Ragge N, Parker MJ, Crow YJ, Kerr B, Kingston H, Metcalfe K, Chandler K, Magee A, Stewart F, McConnell VPM, Donnelly DE, Berland S, Houge G, Morton JE, Oley C, Revencu N, Park S, Davies SJ, Fry AE, Lynch SA, Gill H, Schweiger S, Lam WWK, Tolmie J, Mohammed SN, Hobson E, Smith A, Blyth M, Bennett C, Vasudevan PC, García-Miñaúr S, Alex Henderson A, Goodship J, Wright MJ, Fisher R, Gibbons R, Price SM, C de Silva D, Temple IK, Collins AL, Lachlan K, Elmslie F, McEntagart MI, Castle B, Clayton-Smith J, Black GC, Donnai D. 2012. How genetically heterogenous is Kabuki syndrome?: *MLL2* testing in 116 patients, review and analyses of mutation and phenotypic spectrum. Eur J Hum Genet 20: 381–388.

Bokinni Y. 2012. Kabuki syndrome revisited. J Hum Genet 57:223–227.

FitzGerald K, Diaz M. 1999. MLL2: A new mammalian member of the *trx/MLL* family of genes. Genomics 59:187–192.

Gabrielli O, Bruni S, Bruschi B, Carloni I, Coppa GV. 2002. Kabuki syndrome and growth hormone deficiency: Description of a case treated by long-term hormone replacement. Clin Dysmorphol 11:71–72.

Genevieve D, Amiel J, Viot G, Le Merrer M, Sanlaville D, Urtizberea A, Gerard M, Munnich A, Cormier-Daire V, Lyonnet S. 2004. Atypical findings in Kabuki syndrome: Report of 8 patients in a series of 20 and review of the literature. Am J Med Genet Part A 129A:64–68.

Hannibal MC, Buckingham KJ, Ng SB, Ming JE, Beck AE, McMillin MJ, Gildersleeve HI, Bigham AW, Tabor HK, Mefford HC, Cook J, Yoshiura K, Matsumoto T, Matsumoto N, Miyake N, Tonoki H, Naritomi K, Kaname T, Nagai T, Ohashi H, Kurosawa K, Hou JW, Ohta T, Liang D, Sudo A, Morris CA, Banka S, Black GC, Clayton-Smith J, Nickerson DA, Zackai EH, Shaikh TH, Donnai D, Niikawa N, Shendure J, Bamshad MJ. 2011. Spectrum of MLL2(ALR) mutations in 110 cases of Kabuki syndrome. Am J Med Genet Part A 155A:1511–1516.

Issaeva I, Zonis Y, Rozovskaia T, Orlovsky K, Croce C, Nakamura T, Mazo A, Eisenbach L, Canaani E. 2007. Knockdown of ALR (MLL2) reveals ALR target genes and leads to alterations in cell adhesion and growth. Mol Cell Biol 27:1889–1903.

Kuroki Y, Suzuki Y, Chyo H, Hata A, Matsui I. 1981. A new malformation syndrome of long palpebral fissures, large ears, depressed nasal tip, and skeletal anomalies associated with postnatal dwarfism and mental retardation. J Pediatr 99:570–573.

Lederer D, Grisart B, Digilio M, Benoit V, Crespin M, Ghariani S, Maystadt I, Dallapiccola B, Verellen-Dumoulin C. 2012. Deletion of KDM6A, a histone demethylase interacting with MLL2, in three patients with Kabuki syndrome. Am J Hum Genet 90:119–124.

Ma KH, Chow SN, Yau FT. 2005. Isolated adrenocorticotropin deficiency in a child with Kabuki syndrome. J Pediatr Endocrinol Metab 18: 607–609.

Matsumoto N, Niikawa N. 2003. Kabuki make-up syndrome: A review. Am J Med Genet Part C 117C:57–65.

Ming JE, Russel KL, McDonald-McGinn DM, Japlan P, Zackai EH. 2001a. Unusual systemic manifestations in Kabuki syndrome: Hypoglycemia, malabsorption, and autoimmune thrombocytopenia and anemia. Am J Hum Genet 69:620.

Miyake N, Mizuno S, Okamoto N, Ohashi H, Shijna M, Ogata K, Tsurusaki Y, Nakashima M, Saitsu H, Niikawa N, Matsumoto N. 2013. KDM6A point mutations cause Kabuki syndrome. Hum Mutat 34:108–110.

Ng S, Bigham A, Buckingham K, Hnnibal M, McMillin MJ, Gildersleeve HI, Beck AE, Tabor HK, Cooper GM, Mefford HC, Lee C, Turner EH, Smith JD, Rieder MJ, Yoshiura K, Matsumoto N, Ohta T, Niikawa N, Nickerson DA, Bamshad MJ, Shendure J. 2010. Exome sequencing identifies MLL2 mutations as a cause of Kabuki syndrome. Nat Genet 42:790–793.

Niikawa N, Matsuura N, Fukushima Y, Ohsawa T, Kajii T. 1981. Kabuki make-up syndrome: A syndrome of mental retardation, unusual facies, large and protruding ears, and postnatal growth deficiency. J Pediatr 99: 565–569.

Niikawa N, Kuroki Y, Kajii T, Matsuura N, Ishikiriyama S, Tonoki H, Ishikawa N, Yamada Y, Fujita M, Umemoto H, Iwama Y, Kondoh I, Fukushima Y, Nako Y, Matsui I, Urakimi T, Aritaki S, Hara M, Sukuki Y, Chyo H, Sugio Y, Hasegawa T, Yamanaka T, Tsukino R, Yoshida A, Nomoto N, Kawahito S, Aihara R, Toyota S, Ieshima A, Funaki H, Ishitobi H, Ogura S, Furumae S, Yoshino M, Tsuji Y, Kondoh T, Matsumoto T, Abe K, Harada N, Miike T, Ohdo S, Naritomi K, Abushwereb AK, Braun OH, Schmid E. 1988. Kabuki makeup (Niikawa-Kuroki) syndrome: A study of 62 patients. Am J Med Genet 31:565–590.

Paulussen AD, Stegmann AP, Blok MJ, Tserpelis D, Posma-Velter C, Detisch Y, Smeets EE, Wagemans A, Schrander JJ, van den Boogaard MJ, van der Smagt J, van Haeringen A, Stolte-Dijkstra I, Kerstjens-Frederikse WS, Mancini GM, Wessels MW, Hennekam RC, Vreeburg M, Geraedts J, de Ravel T, Fryns JP, Smeets HJ, Devriendt K, Schrander-Stumpel CT. 2011. MLL2 mutation spectrum in 45 patients with Kabuki syndrome. Hum Mutat 32:E2018–E2025.

Schrandr-Stumpel C, Spruyt L, Curfs L, Defloor T, Schrander J. 2005. Kabuki syndrome: Clinical data in 20 patients, literature review and further guidelines for preventive management. Am J Med Genet Part A 132A:234–243.