

## Molecular Characterization and Management of Congenital Hyperinsulinism: A Tertiary Centre Experience

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**Background:** There is limited data from India regarding medical management of congenital hyperinsulinism (CHI).

**Objective:** To study the molecular diagnosis, medical management and outcomes of children with CHI.

**Study design:** Ambispective.

**Participants:** Children with CHI admitted in from December, 2011 till March, 2020 at a tertiary care referral hospital.

**Outcomes:** Clinical and genetic profile, treatment, and response

**Results:** 42 children with a median age of 3 days (range 1 day to 6 years) were enrolled, of which 23 (54.7%) were diazoxide-responsive. Mutations were identified in 28 out of 41 (68.2%) patients. The commonest gene affected was *ABCC8* in 22

patients. The pathogenic variant c.331G>A in *ABCC8* gene was identified in 6 unrelated cases from one community. Good response to daily octreotide was seen in 13 of the 19 (68.4%) diazoxide-unresponsive patients. Monthly long-acting octreotide was initiated and daily octreotide could be stopped or tapered in 9 patients. Sirolimus was tried with variable response in 6 patients but was discontinued in 5 due to adverse effects. Four patients had focal CHI, of which one underwent partial pancreatic resection. The disease severity reduced with age and neurodevelopment was good in the patients with identifiable genetic defects who were optimally managed. **Conclusions:** Medical management of CHI is effective, if compliance can be ensured, with good quality of life and neurological outcomes.

**Keywords:** *ABCC8* gene, Diazoxide, Hypoglycemia, Octreotide, Sirolimus

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Congenital hyperinsulinism (CHI) results from inappropriate release of insulin from the pancreatic  $\beta$ -cells that causes persistent hypoglycemia in neonates and infants, with possible adverse neurodevelopmental outcomes [1,2]. It is a rare disorder with an incidence varying from 1 in 28 000-50 000 births in Western countries to 1 in 2 500 in the Middle East [3,4]. Mutations in around 15 key genes involved in pancreatic insulin secretion have been identified to cause CHI, the most common being *ABCC8* and *KCNJ11* genes, encoding the sulphonylurea receptor (SUR1) and K<sup>+</sup> inward rectifying (KIR6.2) subunits of the K<sub>ATP</sub> channel, respectively [3]. Identification of the underlying genetic etiology and the mode of inheritance is essential for management and counseling regarding the risk of recurrence.

Histopathologically, CHI can be categorized into diffuse or focal forms [5,6]. The focal forms are amenable to cure by resection of the lesion or partial pancreatectomy. In diffuse

CHI, medical treatment with diazoxide and octreotide is the mainstay [7]; though, surgical modality was used in the past in non-responders [8]. In recent years, medical management with the use of newer options such as long-acting formulations of octreotide have emerged as the treatment of choice [7]. There is limited data from India regarding medical management and outcome of infants with CHI [9,10]. In this study, we present our long-term experience in the management of infants with CHI in India.

### METHODS

This was an ambispective study of the clinical profile, biochemical and molecular diagnoses, drug response and follow-up assessment of children with a diagnosis of CHI, who were admitted or referred to the Pediatric Endocrine Division of the Department of Pediatrics at a tertiary care hospital in the last nine years between December, 2011, when genetic testing was incorporated in the CHI

management protocol, to March, 2020. Patient data was retrieved from records and also collected prospectively from 2017 onwards. Patients not under regular follow-up were contacted via telephone or email for collecting information on their current treatment, growth and development. The study was approved by the ethics committee of our institution.

The diagnosis of CHI was based on recurrent hypoglycemia, glucose requirement  $>8$  mg/kg/min to maintain euglycemia in the newborn period, critical sample serum insulin of  $>2$  mIU/L, inappropriately suppressed blood ketone ( $<2$  mmol/L) or negative urinary ketone, and inappropriate glycemic response to glucagon challenge test (GCT). Other investigations included critical sample for growth hormone (GH), cortisol, lactate, galactosemia screen and serum ammonia levels.

Genetic testing of the proband (along with the parents in most cases) was performed either at Department of Molecular Genetics, Royal Devon and Exeter NHS Foundation Trust, Exeter, UK or Department of Molecular Genetics, Madras Diabetes Research Foundation (MDRF), Chennai after taking informed consent from the parents. Sanger sequencing was performed for *ABCC8* and *KCNJ11* genes first, and for *GLUD1*, *GCK*, *HADH* in patients with specific suggestive features. Targeted next generation sequencing (tNGS) of the coding regions and exon/intron boundaries of known CHI genes (panel consisting of *KCNJ11*, *ABCC8*, *GLUD1*, *GCK*, *HADH*, *HNF4A*, *INSR*, *SLC16A1*, *TRMT10A* and *HNF1A*) was obtained (Agilent custom capture v5.3/Illumina NextSeq500) in patients with persistent diazoxide-unresponsive hypoglycemia in whom no pathogenic variants were identified in the common genes. Nuclear scan (18-F DOPA PET) was performed in-house to rule out focal pancreatic pathology where indicated.

Oral diazoxide was used as the first line of treatment in a dose of 5-15 mg/kg/day in 3 divided doses. Hydrochlorothiazide was added at a dose of 1.5-2.5 mg/kg/day to prevent fluid overload. The patient was deemed diazoxide-responsive if the blood glucose normalized and glucose infusion could be tapered off. Minimum of 5 days of treatment at a maximum dose of 20 mg/kg/day was given before labeling the patient as diazoxide-unresponsive. The second line of treatment consisted of injection octreotide with a starting dose of 15-20  $\mu$ g/kg/day (maximum 50  $\mu$ g/kg/day). In case of sub-optimal response to octreotide, a trial of sirolimus was given in a dose of 0.05-1.8 mg/m<sup>2</sup> to keep the plasma drug level between 5-15 ng/mL with monitoring of side effects. Where feasible, patients on long-term octreotide were switched to long-acting release (LAR) octreotide intramuscularly monthly, at a dose calculated as daily dose

multiplied by 30. After starting monthly injections, daily octreotide was tapered and stopped over the next few months, where feasible. A blood glucose value  $>70$  mg/dL was considered normal. The treatment was considered effective if 90% blood sugar values were normal with no severe hypoglycemia ( $<30$  mg/dL) or hypoglycemic seizures.

Most of the enrolled subjects were a few weeks old at the time of hospitalization. They were managed with frequent 2-3 hourly feeds (expressed breastmilk or formula) orally or via nasogastric tube. Feeds were supplemented with uncooked cornstarch at 1-2 g/kg/day, or with glucose 0.5 g/kg/feed, as needed.

Importance of compliance with medication and regular feeding, especially nocturnal feeds, was emphasized as part of education at discharge. Parents were taught to administer tube feeds, monitor blood glucose, manage hypoglycemia, and give subcutaneous octreotide (where indicated). Post-weaning, a balanced diet with avoidance of foods with high glycemic index was advised.

Patients were followed up monthly in the first year of life. Anthropometry and development assessment were performed in each follow-up visit. Children with delayed milestones with or without microcephaly were advised formal developmental assessment, sensory stimulation and magnetic resonance imaging (MRI) brain. Those on octreotide were monitored with liver function tests every three months and ultrasound abdomen for gall bladder stones every six months.

*Statistical analysis:* Statistical analysis was carried out using STATA version 14.0. Continuous variables were compared using student *t* test and proportions with chi-square test. A *P* value  $<0.05$  was considered statistically significant

## RESULTS

A total of 42 patients (23 males) were included in the study, 26 of whom had presented in the neonatal period. The median age of presentation was day 3 (range 1 day to 6 years). Consanguinity was present in 4 families. Two families had two affected siblings and three families had history of previous neonatal deaths. Hypoglycemic seizure was the most common presentation, and macrosomia was present in 15/26 (57.7%) neonates indicating in utero hyperinsulinism.

Mutations were identified in 28 (68.2%) patients, with *ABCC8* gene mutations in 22 (53.6%) patients. Of these, 14 had autosomal recessive mode of inheritance (homozygous and compound heterozygous in 8 and 6 patients, respectively), six had a paternally inherited heterozygous mutation, and two had de novo autosomal dominant mutations.

Twenty-three (54.7%) patients responded to diazoxide. Mutations could be identified in 9/22 (41%) of the diazoxide-responsive cases (1 could not be tested) and all the diazoxide-unresponsive patients. **Table I** presents the clinical comparison between diazoxide-responsive and unresponsive patients.

**Web Table I** summarizes the clinical presentation, genetic defects, F-DOPA PET results, treatment and follow-up of the patients. A total of 22 different pathogenic/likely pathogenic variants were identified in *ABCC8* gene, of which three variants were novel, namely c.3653+2T>G (abnormal splicing), c.2423\_2424del (p.L808fs), and c.4411G>A (p.D1471N). All the novel variants were predicted to be pathogenic using prediction software, and were present in heterozygous state in clinically unaffected parents. The pathogenic variant c.331G>A (p.G111R) was common, and present in six patients; in homozygous state in two (cases 8, 14), compound heterozygous in three (cases 1, 3 and 13) and paternally-inherited heterozygous in one (case 20).

Two babies (patient 41 and 42) with prematurity and intrauterine growth retardation had transient but prolonged hyperinsulinism and succumbed at 1 month and 3 months of age, with prematurity and liver failure, respectively. One baby had no identifiable genetic mutation while the other could not be tested.

Subcutaneous octreotide was started in 19 patients

**Table I Response to Diazoxide in Children With Congenital Hyperinsulinism (N=42)**

Characteristic	Diazoxide responsive (n=23)	Diazoxide unresponsive (n=19)
Male gender	10 (43.5)	11 (57.8)
Preterm birth	3 (13)	4 (21)
Birthweight <sup>b,c</sup>	2.86 (0.68)	3.61 (0.69)
Large for gestational age <sup>c</sup>	2 (8.7)	14 (73.6)
Neonatal onset <sup>c</sup>	7 (30.4)	19 (100)
Onset of hypoglycemia <sup>a,c</sup>	3 mo (1 d-6 y)	1 d (1-10 d)
Age of referral <sup>a,c</sup>	7 mo (1 mo-12 y)	1 mo (12 d-5 mo)
Genetic mutations <sup>c</sup>	9/22 (41)	19 (100)
Pathogenic variants	<i>ABCC8</i> (paternal), 1; <i>ABCC8</i> (AD), 2; <i>HADH</i> (AR), 2; <i>GLUDI</i> (AD), 3; <i>KMD6A</i> , 1	<i>ABCC8</i> (AR), 14; <i>ABCC8</i> (paternal), 5
Neurodevelopmental issues	12/20 (60)	3/17 (17.6)

Data expressed as n (%) or <sup>a</sup>median (range) or <sup>b</sup>mean (SD). AD: autosomal dominant, AR: autosomal recessive; neurodevelopmental issues - developmental delay or behavioral problems. <sup>c</sup>P < 0.05.

with diazoxide-unresponsive disease (autosomal recessive or paternally inherited heterozygous pathogenic variants in *ABCC8*). Thirteen patients achieved good response at a median daily dose of 40 (range 35-50) µg/kg/day, while 4 continued to have hypoglycemia. Nine patients were started on monthly LAR octreotide injection at the median age of 6 month (range 3 month - 4 year). Daily octreotide was gradually stopped over the next 3-12 months in five patients, and the doses were reduced in remaining four patients.

Sirolimus was started in six patients with partial response. However, it was discontinued in one patient because of inability to reach therapeutic blood levels on maximum dose, and in four other patients after a variable duration of treatment due to adverse effects. None of our patients received long term glucagon treatment.

Six patients had paternally inherited heterozygous *ABCC8* mutations (cases 15-20); three out of four patients had focal disease on I 8-F DOPA-PET, Of these, one patient (case 19) with a focal lesion at the junction of tail and body of pancreas underwent partial pancreatic resection at 7 months of age and has remained euglycemic till date. Second patient (case 16) was well controlled on octreotide and the treatment was stopped at 1 year of age. The third patient (case 17) had a lesion in the head and uncinate process of pancreas, which was deemed inoperable. She was continued on octreotide and sirolimus with few episodes of hypoglycemia related to poor compliance.

Hypertrichosis to a variable degree was noted in all infants receiving diazoxide, which reversed partially as the dose decreased on follow-up. One patient (case 6) developed congestive heart failure at initial trial. Case 39 developed neutropenia after 3 months of treatment and diazoxide had to be discontinued. One baby (case 42) with IUGR developed jaundice and liver failure after starting diazoxide.

Five patients developed gastrointestinal intolerance with octreotide. One patient had constipation and rectal bleeding with octreotide given at 55 µg/kg/day, and the dose had to be decreased. Transient mild elevation of transaminases was noted in three patients, but none had significant derangement needing withdrawal of the drug. One patient had asymptomatic gallstones and biliary sludge that improved with UDCA.

A derangement in liver function was seen in two patients on sirolimus. One patient (case 4) was euglycemic on sirolimus monotherapy, but developed life-threatening sepsis with shock at 4 years of age after which sirolimus was switched to LAR octreotide. Case 3 developed refractory anemia not responding to iron after which sirolimus was stopped. Case 17 was lost to follow-up during the Covid-19

lockdown and died at 1.7 years due to sepsis in another hospital.

The median age till follow-up was 3.9 year (range 3 month - 15 year) and 3.5 years (range 3 month - 9 year) in diazoxide-responsive and unresponsive disease, respectively. On follow-up, the growth parameters of all patients on diazoxide and most patients on octreotide were within normal range. Two patients on octreotide had height <-3SD with short mid-parental height where causal role of octreotide with short stature could not be established. Case 3, with height at -3.8 SD at 3 year showed improvement to -3.1 SD over the next 1.5 years after stopping octreotide. Three patients developed obesity in infancy. Of the 14 patients with autosomal recessive *ABCC8* mutations (P1-14), two children had died, seven had normal development, three had initial mild motor delay (which improved with time), and two had global developmental delay. Of note, 9 out of 11 alive patients had normal neurodevelopment in follow up. Among the 6 patients with paternally inherited heterozygous *ABCC8* mutations, five had normal neuro-development, while one had frequent hypoglycemic seizures with global delay, and died at 1.7 year. Among the diazoxide-responsive cases, the neurodevelopmental outcomes were highly variable.

## DISCUSSION

In this series of children with CHI from India, mutations could be identified in the majority, the commonest being autosomal recessive *ABCC8*, similar to the observations in previous Indian data [9]. The response rate to diazoxide in our study was similar to the reported rate of 50-65% [11,12]. The positivity rate of mutations in this study was similar to the previously reported rate, suggesting that the etiology of diazoxide-responsive CHI was very heterogeneous and not fully elucidated [11,12].

All the six patients with the pathogenic variant c.331G>A (p.G111R) in *ABCC8* gene, although unrelated and from different states of Northern India, belonged to the Aggarwal community. This is interesting as caste endogamy is prevalent in this community and it is known to harbor founder mutations for other rare autosomal recessive disorders such as panthothenate kinase associated neuro-degeneration, and megalencephalic-leukodystrophy with cysts [13].

Our observations suggest that medical therapy can be used in focal disease if euglycemia is achieved on a single drug, as there are chances of spontaneous remission. Non-availability of surgical expertise and family's preference are also relative indications of medical management in focal CHI [14]. 18-F DOPA-PET is highly useful for localizing focal lesions [15]; however, its availability is limited.

Majority of patients with homozygous *ABCC8* mutations and diffuse CHI responded well to octreotide, in consonance with previous literature [14,16], and nine of these patients were shifted to long-acting formulations. Lanreotide and sandostatin-LAR are two long-acting somatostatin analogues [17], of which only the latter is available in India. It is used in parallel with daily octreotide therapy for few months, till blood levels of octreotide reach adequate levels [18]. There are limited reports on use of long-acting octreotide in CHI suggesting better glycemic control [18,19].

Diazoxide can lead to multiple side effects including pulmonary hypertension (PH) [7,20]. PH is reported in 2.4-7% patients, especially in children with congenital heart disease [21], but we did not routinely monitor for this. Long-acting octreotide formulations should be used only beyond the neonatal period. Suppression of growth hormone and growth failure are occasionally reported with somatostatin analogues, but catch-up growth occurs in follow-up after octreotide is weaned [14,16], as was observed in one of our patients. Octreotide therapy can lead to suppression of thyroid stimulating hormone to recommend regular monitoring of thyroid function [7], though none of our patients had deranged thyroid function on follow-up. Sirolimus, an mTOR inhibitor, has been reported to reduce the proliferation of pancreatic  $\beta$ -cells and inhibit insulin secretion [22], was found to be effective in conjunction with octreotide in two patients, and as monotherapy in one. However, it had to be discontinued in most patients due to serious side effects; as also reported recently in a follow-up study in 22 patients [23].

There were multiple challenges in the medical management, the foremost being cost of daily octreotide and LAR preparations. Lack of free availability of diazoxide, frequent feeding and monitoring of blood sugar, and compliance to multiple daily injections were additional challenges faced by the families. The use of LAR octreotide helped in improving compliance to treatment. Non-response to diazoxide and post-surgical diabetes requiring insulin therapy have been identified as major drivers of cost in a previous study [24]. Medical therapy although expensive in the short-term, can help reduce overall costs as clinical disease remits with age.

Neurodevelopmental outcomes were highly variable in diazoxide-responsive patients, depending on age of diagnosis and referral, underlying molecular mechanism and compliance to therapy. Overall, in our study, developmental outcomes were poorer in those with low compliance or delayed diagnosis. Hypotonia, fine motor problems, clumsiness and speech problems were reported in medically treated diazoxide-unresponsive children,

which resolved by 4-5 years of age with subsequent normal neurodevelopment [14].

The limitations of our study are that compliance could not be formally documented in all patients, and detailed neurodevelopmental assessment for subtle issues in behavior, learning, attention and speech was not done in all patients.

To conclude, in this series of medically treated Indian children with CHI, the main challenges were related to frequent monitoring, feeding, compliance to medication and high cost of therapy. Good neurodevelopmental outcomes were observed in those with optimal care and appropriate medical therapy. Remission or reduction in severity after the first two years of life was noted, which is a silver lining in the management of this difficult disease.

*Ethics clearance:* Ethics committee of AIIMS, New Delhi; No. IEC 109/5.2.21, RP-26/2021, dated February 05, 2021.

*Contributors:* RS, KR: prepared the manuscript; RS, KR, AS, PMN, AK, ND, VJ: involved the diagnostic work up, clinical management of patients and data collection; SEF, JALH, VR, VM: performed the genetic studies; VJ: conceived the study, initiated the collaborations for genetic testing, critically reviewed the manuscript. All authors approved the final manuscript and are accountable for all aspects of the work.

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*Competing interests:* None stated.

Note: Additional material related to this study is available with the online version at [www.indianpediatrics.net](http://www.indianpediatrics.net)

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**Web Table I: Summary of Genetic Mutations, Treatment and Follow-Up in Patients with Congenital Hyperinsulinism (N=40)**

S. no.	Age at onset	Pathogenic variant				F-18 DOPA PET	Initial treatment	Follow-up			
		Gene, zygosity	Nucleotide change	Protein change	Type of variant			Age	Treatment	Growth	Development
P1	Day 8	ABCC8, Compound hetero	c.331G>A/ c.3979G>T	p.G111R/p.E1327*	Missense/ Nonsense	Diffuse	Octreotide 35 µg/kg/day LAR started at 3 y	9 y	LAR 7.5 mg every 45 days	Normal	Initial motor delay Average school performance
P2	Day 1	ABCC8, Compound hetero	c.1330C>T/ c.3653+2T>A	p.Q444*/p.?	Nonsense/ Aberrant splicing variant^	Diffuse	Octreotide 40 µg/kg/day LAR 10 mg per month from 2.5 y, stopped at 3.5 y	4.5 y	None	Normal	Mild motor delay Delayed speech
P3	Day 1	ABCC8, Compound hetero	c.221G>A/ c.331G>A	p.R74Q and p.G111R	Missense/ Missense	Diffuse	Octreotide 50 µg/kg/day; Sirolimus added but stopped due to severe anemia. Octreotide tapered and stopped at 2 y	4.5 y	None	Ht -3 SD Rest normal	Normal
P4	Day 2	ABCC8, Homo	c.522dup/ c.522dup	p.L175fs/p.L175fs	Frameshift	Diffuse	Poor response and compliance to octreotide Sirolimus 1.8 mg/m <sup>2</sup> Developed sepsis at 4 y, shifted to octreotide LAR	4 y	LAR 10 mg monthly	Normal	GDD, DQ 70, epilepsy on AED
P5	Day 1	ABCC8, Compound hetero	c.4480C>T/ c.(4414+1_4414-1)_(*4749+34 ?) del	p.R1494W/p.?	Missense/ Deletion	Not done	Octreotide 40 µg/kg/day	Lost to FU after 6 mo	-	Normal	Global delay
P6	Day 1	ABCC8, Homo	c.3653+2T>G/ c.3653+2T>G	p.*/p.?	Aberrant splicing^	Diffuse	Octreotide 40 µg/kg/day LAR 12 mg monthly since 2 y of age Sirolimus trial; stopped due to low plasma drug levels	3.5 y	Daily octreotide at 10 µg/kg/day LAR 12 mg monthly	Height -3.5 SD Rest normal	Initial mild motor delay Normal in FU
P7	Day 1	ABCC8, Homo	c.2423_2424del/ c.2423_2424del	p.L808fs/p.L808fs	Frameshift^	Diffuse	Octreotide 35 µg/kg/day LAR 10 mg monthly since 6 mo of age	2.5 yrs	LAR 10 mg monthly	Height -2 SD	Normal
P8	Day 1	ABCC8, Homo	c.331G>A/ c.331G>A	p.G111R/p.G111R	Missense	Diffuse	Octreotide 35 µg/kg/day. Sirolimus tried but stopped due to deranged LFT. LAR 5 mg monthly started at 4 mo, daily octreotide stopped after 1.5 y.	2.5 yrs	LAR 10 mg q 2 monthly	Weight for height 97 <sup>th</sup> centile Rest normal	Normal, h/o myoclonic jerks controlled on AED
P9	Day 1	ABCC8, Homo	c.3653+2T>G/ c.3653+2T>G	p.*/p.?	Aberrant splicing^	Diffuse	Octreotide 40 µg/kg/day. Sirolimus tried but stopped due to deranged LFT. LAR 10 mg monthly started at	2 yrs	LAR 10 mg monthly	Obese Wt for height 99 <sup>th</sup> centile Rest normal	Normal

							4 months of age				
<b>P10</b>	Day 1	ABCC8, Homo	c.2675_2679del/c.2675_2679del	p.Q892fs/p.Q892fs	Frameshift	Not done	Octreotide 50 µg/kg/day Planned for near-total pancreatectomy	LAMA, expired	-	-	-
<b>P11</b>	Day 2	ABCC8, Compound hetero	c.267delT and c.619-629 del CCCGAGG ACCT	p.(Ile89MetfsTer10)/p.(Pro207AlafsTer61)	Frameshift	Not done	Octreotide 40 µg/kg/day	Expired at 4 months due to hypoglycemia at night	-	-	-
<b>P12</b>	Day 1	ABCC8, Homo	c.1138del	p.(Ala380fs)/p.(Ala380fs)	Frameshift	Diffuse	Octreotide 40 µg/kg/day LAR started at 5 months of age	1 y	LAR 10 mg monthly Octreotide 10 µg/kg/day	Wt for Ht + 2SD Rest normal	Normal
<b>P13</b>	Day 1	ABCC8, Compound hetero	c.331G>A/c.4411G>A	p.Gly111Arg/p.Asp147Asn	Missense/Missense^	Diffuse	Octreotide 50 µg/kg/day LAR 5 mg monthly started at 2 mo	4 mo	Octreotide 40 µg/kg/day, LAR 5 mg monthly	Normal	Normal
<b>P14</b>	Day 2	ABCC8, Homo	c.331G>A/c.331G>A	p.G111R/p.G111R	Missense	Not done	Octreotide 40 µg/kg/day	3 mo	Octreotide 40 µg/kg/day	Normal	Normal
<b>P15</b>	Day 1	ABCC8, Paternally inherited hetero	c.3871-1G>A	p.?/N	Aberrant splicing	Diffuse	Octreotide 10 µg/kg/day	9 mo	LAR 2 mg monthly	Normal	Normal
<b>P16</b>	Day 1	ABCC8, Paternally inherited hetero	c.4024C>T/N	p.Q1342*/N	Missense	Focal	Octreotide 10 µg/kg/day	2.5 y	Off treatment since 1.2 y	Normal	Normal
<b>P17</b>	Day 1	ABCC8, Paternally inherited hetero	c.4256G>A/N	p.R1419H/N	Missense	Focal	Octreotide 40 µg/kg/day Sirolimus 0.3 mg/m <sup>2</sup>	1.5 y Lost to FU: Expired at 1.7 years due to sepsis	Octreotide 40 µg/kg/day sirolimus Poor compliance	Normal	Moderate GDD
<b>P18</b>	Day 10	ABCC8, Paternally inherited hetero	c.1330C>T	p.Q444*/N	Nonsense	Not done	Octreotide 20 µg/kg/day till 2 years of age	7 y	Off treatment since 2 years of age	Normal	Normal
<b>P19</b>	Day 3	ABCC8, Paternally inherited hetero	c.4415-13G>A	p.?/N	Aberrant splicing	Focal	Octreotide 12 µg/kg/day Partial pancreatectomy done at 7 months of age	3 y	None	Normal	Normal
<b>P20</b>	Day 6	ABCC8, Paternally inherited hetero	c.331G>A/N	p.G111R	Missense	Not done	Diazoxide	4.5 y	4 mg/kg	Normal	Normal
<b>P21</b>	Day 3	ABCC8, Hetero (de novo)	c.4377G>C/N	p.Q1459H	Missense	No focal lesion	Diazoxide	8 y	Stopped after 6 years	Normal	Mild MR
<b>P22*</b>	Day 1	ABCC8, Hetero (de novo)	c.4519G>A	p.Glu1507Lys	Missense	Not done	Diazoxide	18 mo	3.5 mg/kg	Normal	Mild GDD

P2 3	3 mo	HADH, Homo	c.550A>T	p.I184F	Missense	Not done	Diazoxide	5 y	5 mg/kg	Normal	Normal
P2 4	4 mo	HADH, Homo	c.550A>T	p.I184F	Missense	Not done	Diazoxide	6.5 y	5 mg/kg	Normal	Mild delay
P2 5	7 mo	GLUD1, Hetero <sup>#</sup>	c.943C>T/N	p.H315 Y	Missense	Not done	Diazoxide	4 y	4 mg/kg	Normal	Autism
P2 6	6 mo	GLUD1, Hetero <sup>#</sup>	c.1334C>T/ N	p.S445L	Missense	Not done	Diazoxide	7 y	7 mg/kg, stopped at 6 yrs due to non- availability Octreotide 15 ug/day	Microce phaly	GDD, Epilepsy
P2 7	4 mo *	GLUD1, <sup>#</sup> Hetero	c.820C > T/N	p.R274 C	Missense	Diffuse	Diazoxide	10 mo	7 mg/kg	Normal	Fine motor delay
P2 8	6 yrs	None by tNGS	-	-	-	Not done	Diazoxide	15 y	2 mg/kg	Obese	-
P2 9	3 mo	None in ABCC8/ KCNJ11	-	-	-	Not done	Diazoxide	Lost to follow- up at 3 y	Poor compliance	-	GDD Epilepsy
P3 0	4 mo	None in ABCC8/ KCNJ11	-	-	-	Not done	Diazoxide	6 y	0.4 mg/kg Poor compliance	Normal	GDD, poorly controlled epilepsy
P3 1	9 mo	None by tNGS	-	-	-	No focal lesion	Diazoxide	6.5 y	5 mg/kg	Normal	GDD, epilepsy
P3 2	6.5 mo	None by tNGS	-	-	-	Not done	Diazoxide	5 y	5 mg/kg	Normal	Normal
P3 3	3 mo	None in ABCC8/KC NJ11	-	-	-	Not done	Diazoxide	Lost to FU	-	-	-
P3 4	5 mo	None in ABCC8/KC NJ11	-	-	-	Not done	Diazoxide	Lost to FU after 3 y, Expired at 4 y due to hypogly cemia	-	-	-
P3 5	9 mo	None by tNGS	-	-	-	Not done	Diazoxide	5 y	2 mg/kg	Normal	Normal
P3 6	Day 6	None by tNGS	-	-	-	Not done	Diazoxide	3 y	4 mg/kg	Normal	GDD, epilepsy
P3 7	Day 3	None by tNGS	-	-	-	Not done	Diazoxide	2.2 y	2.5 mg/kg	Normal	Mild GDD, epilepsy
P3 8	5 mo	None by tNGS	-	-	-	Not done	Diazoxide	1.5 y	10 mg/kg	Normal	GDD, epilepsy
P3 9	2 mo	KMD6A mutation causing Kabuki syndrome Mosaic	ChrX:g.(?_4 4873552) (44873987_ ?)del	De novo	-	Not done	Diazoxide, stopped due to neutropenia, Started Octreotide	Lost to FU after 1 y	Octreotide 20 ug/kg/day	Length < -2SD	GDD, epilepsy
P4 0	2 yr	None by tNGS					Diazoxide	8 y	1.5 mg/kg	Normal	Normal

Abbreviations: Homo: homozygous; Hetero: heterozygous; tNGS: Targeted next generation sequencing; LAR: Long-acting release octreotide; FU: follow-up; LAMA: Left against medical advice; Wt: weight; Ht: height, SD: Standard deviations, AED: antiepileptic drugs; DQ: Development quotient; GDD: global developmental delay; \*One affected elder sibling diagnosed at 5 years of age and having epilepsy; \*\*sibling of 23; #Hyperinsulinism-hyperammonemia syndrome; ^Novel pathogenic/ likely pathogenic variants