Mechanisms of Disease: advances in diagnosis and treatment of hyperinsulinism in neonates

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SUMMARY

Hyperinsulinism is the single most common mechanism of hypoglycemia in neonates. Dysregulated insulin secretion is responsible for the transient and prolonged forms of neonatal hypoglycemia, and congenital genetic disorders of insulin regulation represent the most common of the permanent disorders of hypoglycemia. Mutations in at least five genes have been associated with congenital hyperinsulinism: they encode glucokinase, glutamate dehydrogenase, the mitochondrial enzyme short-chain 3-hydroxyacyl-CoA dehydrogenase, and the two components (sulfonylurea receptor 1 and potassium inward rectifying channel, subfamily J, member 11) of the ATP-sensitive potassium channels (K_{ATP} channels). K_{ATP} hyperinsulinism is the most common and severe form of congenital hyperinsulinism. Infants suffering from K_{ATP} hyperinsulinism present shortly after birth with severe and persistent hypoglycemia, and the majority are unresponsive to medical therapy, thus requiring pancreatectomy. In up to 40–60% of the children with K_{ATP} hyperinsulinism, the defect is limited to a focal lesion in the pancreas. In these children, local resection results in cure with avoidance of the complications inherent to a near-total pancreatectomy. Hyperinsulinism can also be part of other disorders such as Beckwith–Wiedemann syndrome and congenital disorders of glycosylation. The diagnosis and management of children with congenital hyperinsulinism requires a multidisciplinary approach to achieve the goal of therapy: prevention of permanent brain damage due to recurrent hypoglycemia.

KEYWORDS ATP-sensitive potassium channels, glucokinase, glutamate dehydrogenase, hyperinsulinism, hypoglycemia

REVIEW CRITERIA

We searched for original articles focusing on hyperinsulinism in PubMed between 1970 and 2006. The search terms we used were "hypoglycemia", "hyperinsulinism", "K_{ATP} channels", "glucokinase", "glutamate dehydrogenase", and "short chain 3-hydroxyacyl-CoA dehydrogenase". All papers identified were English-language full text papers. We also searched the references lists of identified articles for further papers.

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INTRODUCTION

Etiology of neonatal hypoglycemia

Hypoglycemia is a frequent problem in newborn infants that must be diagnosed and treated efficiently to avoid seizures and permanent brain damage. Management is complicated by the fact that neonates present a remarkably wide range of possible causes for hypoglycemia. These include transient forms of hypoglycemia that can occur in normal infants, more prolonged forms of hypoglycemia that are associated with complications of gestation and delivery, and neonatal presentation of permanent hypoglycemia disorders due to endocrine or metabolic diseases (Box 1).

The risks of these various forms change dramatically over the first few days after birth. For example, it has been known for over 30 years that up to 50% of normal neonates are unable to maintain plasma glucose above 50 mg/dl with fasting for as little as 8 h after delivery.[1] The high risk of hypoglycemia in normal neonates reflects incomplete development at birth of the pathways for hepatic gluconeogenesis and ketogenesis. These pathways quickly mature and by 2–3 days of life, the susceptibility of normal infants to hypoglycemia resolves. Maternal diabetes or medications may be important contributors to the risk of hypoglycemia during the first few days after delivery. Peripartum complications, such as birth asphyxia or other perinatal stresses, can be associated with prolonged hypoglycemia that persists for several weeks after birth.

As shown in Box 1, with the exception of the normal immaturity of fasting adaptation, which accounts for the high risk of hypoglycemia in the first 12 h, hyperinsulinism is the single most common of the transient and permanent disorders of hypoglycemia. This Review will therefore focus on our current understanding of the various forms of hyperinsulinism in the newborn and their diagnosis and management. Particular emphasis is placed on the genetic disorders associated with congenital hyperinsulinism and

Box 1 The etiology of hypoglycemia in neonates.

Transient neonatal hypoglycemia

Day 1 of life: developmental immaturity of fasting adaptation (mechanism: impaired ketogenesis and gluconeogenesis)

First 2 days of life: transient hypoglycemia due to maternal factors

- maternal diabetes (mechanism: hyperinsulinism)
- intravenous glucose administration during labor and delivery (mechanism: hyperinsulinism)
- medications: oral hypoglycemics, terbutaline, propranolol (mechanism: hyperinsulinism)

Prolonged neonatal hypoglycemia

Perinatal stress-induced hyperinsulinism (low birth weight, birth asphyxia, maternal toxemia or pre-eclampsia, prematurity)

Beckwith–Wiedemann syndrome

Hypopituitarism

Permanent neonatal hypoglycemia (caused by congenital endocrine or metabolic disorders)

Congenital hyperinsulinism

- ATP-sensitive potassium channel hyperinsulinism
- glutamate dehydrogenase hyperinsulinism
- glucokinase hyperinsulinism
- short-chain 3-hydroxyacyl-CoA dehydrogenase hyperinsulinism
- congenital disorders of glycosylation

Counter-regulatory hormone deficiency

- hypopituitarism
- adrenal insufficiency

Gluconeogenesis or glycogenolysis enzyme defects

Fatty acid oxidation disorders

the very common, but as yet poorly understood, prolonged hyperinsulinism associated with perinatal stresses. Other well-known risk factors for neonatal hypoglycemia, such as maternal diabetes or hypoglycemic medications, will not be addressed.

Congenital hyperinsulinism

Congenital hyperinsulinism (CHI) represents a group of clinically, genetically, and morphologically heterogeneous disorders characterized by dysregulated insulin secretion and resulting in severe and persistent hypoglycemia. During the last 10 years, advances in molecular genetics and in the understanding of β-cell biochemical pathways have resulted in the understanding of the pathophysiology of most forms of CHI. Despite these advances, treatment of the infant with CHI continues to be one of the major challenges in pediatric endocrinology.

First described in 1954 by MacQuarrie[2] as "idiopathic hypoglycemia of infancy", CHI has been referred to by a large number of terms, including nesidioblastosis, leucine-sensitive hypoglycemia, islet dysregulation syndrome, and persistent hyperinsulinemic hypoglycemia of infancy. We prefer to denote the condition as hyperinsulinism for simplicity and to avoid confusion, and to refer to the group of permanent inborn conditions as CHI.

Worldwide, CHI occurs at a frequency of 1 in 30,000 to 1 in 50,000 live births,[3] but in some isolated populations rates of 1 in 3,200 have been reported.[4] Higher rates of 1 in 2,500 live births have been reported in areas of high consanguinity such as the Arabian Peninsula.[5]

MOLECULAR BASIS OF CONGENITAL HYPERINSULINISM

Mutations in five proteins have been associated with CHI: the sulfonylurea receptor 1 (SUR-1, a member of the superfamily of ATP-binding cassette proteins, encoded by *ABCC8* [ATP-binding cassette subfamily C, member 8]);[5] Kir6.2 (encoded by *KCNJ11* [potassium inward rectifying channel, subfamily J, member 11]);[6] glucokinase (encoded by *GCK*);[7] glutamate dehydrogenase (GDH; encoded by *GLUD-1* [glutamate dehydrogenase 1]);[8] and the mitochondrial enzyme short-chain 3-hydroxyacyl-CoA dehydrogenase (SCHAD; encoded by *HADH* [hydroxyacyl-coenzyme A dehydrogenase]; formerly *HADHSC*).[9] SUR-1 and Kir6.2 combine to form the β-cell ATP-sensitive potassium channel (K_{ATP} channel). Table 1 summarizes the major features of these disorders.

ATP-sensitive potassium channel congenital hyperinsulinism

Pathophysiology

Loss-of-function mutations in the K_{ATP} channel result in K_{ATP} channel hyperinsulinism (K_{ATP}-HI)—the most common and severe form of CHI. The β-cell K_{ATP} channel is a hetero-octameric complex comprising two subunits: a K^+-selective pore-forming subunit, Kir6.2, and a regulatory subunit, SUR-1. Four Kir6.2 subunits form the central pore, coupled to four SUR-1 subunits. The K_{ATP} channel is inhibited by sulfonylurea drugs (used therapeutically to stimulate insulin secretion in type 2 diabetes) and activated by diazoxide (the main medical treatment for CHI). The K_{ATP} channels couple the energy state of the β-cell to membrane potential by sensing

Table 1 Classification of genetic forms of congenital hyperinsulinism.

Genetic form	Gene	Chromosome	Inheritance	Clinical features	Treatment
K_{ATP}-HI	*ABCC8* *KCNJ11*	11p15	Diffuse: AR Focal: loss of heterozygosity with paternal mutation	Severe, unresponsive to medical therapy	Pancreatectomy
Dominant K_{ATP}-HI	*ABCC8* *KCNJ11*	11p15	AD	Milder, responsive to medical therapy	Diazoxide
GDH-HI	*GLUD-1*	10q	AD	Protein sensitivity, hyperammonemia	Diazoxide
GK-HI	*GCK*	7p	AD	Varies	Diazoxide
SCHAD-HI	*HADH*	4q	AR	Abnormal acyl-carnitine profile	Diazoxide

Abbreviations: *ABCC8*, ATP-binding cassette subfamily C, member 8; AD, autosomal dominant; AR, autosomal recessive; GDH-HI, glutamate dehydrogenase hyperinsulinism; GK-HI, glucokinase hyperinsulinism; *GLUD-1*, glutamate dehydrogenase 1; *HADH,* hydroxyacyl-coenzyme A dehydrogenase; K_{ATP}-HI, ATP-sensitive potassium channel hyperinsulinism; *KCNJ11*, potassium inward rectifying channel, subfamily J, member 11; SCHAD-HI, short-chain 3-hydroxyacyl-CoA dehydrogenase hyperinsulinism.

changes in intracellular phosphate potential (the ATP:ADP ratio; Figure 1). Electrophysiological studies of islets from infants with K_{ATP}-HI show reduction of K_{ATP} channel activity and spontaneously active voltage-dependent Ca^{2+} channels (reviewed by Dunne *et al.*[10]).

Phenotype

K_{ATP}-HI is characterized in most cases by large for gestational age birth weight (because of stimulation of fetal growth by excessive insulin *in utero*), neonatal onset of hypoglycemia, and unresponsiveness to diazoxide therapy. With some exceptions, infants with K_{ATP}-HI fail to respond to medical therapy and require pancreatectomy to control the hypoglycemia.

Genetics of K_{ATP}-HI

The functional regulation of K_{ATP} channels induced by changes in the ATP:ADP ratio involves cooperative interactions of nucleotides at both subunits. Kir6.2 is a weak inward rectifier K^+ channel, comprising 390 amino acids, with two predicted α-helical transmembrane domains. ATP-induced channel closure seems to involve the cytoplasmic domains, although mutations at other sites can also abolish ATP actions. Human SUR-1 consists of 1,581 amino acids and has 17 predicted transmembrane regions (organized into three domains: TMD0, TMD1, and TMD2), 9 cytoplasmic loops (CL1–CL9), and 2 intracellularly disposed nucleotide-binding folds (NBF). Nucleotide hydrolysis is possible at both NBF sites (reviewed by Dunne *et al.*[10]).

The SUR-1 gene (*ABCC8*) comprises 39 exons and is clustered with the Kir6.2 gene (*KCNJ11*), a single open reading frame lying 3' of *ABCC8*. More than 100 mutations of *ABCC8* and 20 mutations of *KCNJ11* have been found. *ABCC8* mutations include splice-site mutations, missense mutations, nonsense mutations, insertions, and deletions, and are spread throughout the coding region of the gene, although about 60% of them cluster in NBF2. Channel activity is completely eliminated by some mutations, whereas others alter the density of the channels or their response to nucleotides (reviewed by Fournet and Junien[11]). Most reported mutations of *ABCC8* and *KCNJ11* are recessive; however, a few dominantly expressed mutations of *ABCC8* and one of *KCNJ11* have been reported.[12–15] The dominant defects retain responsiveness to diazoxide and tend to be milder. Interestingly, heterozygous gain-of-function mutations in *KCNJ11* cause transient or permanent neonatal diabetes.[16]

Histologic forms of K_{ATP}-HI

There are two distinct histological forms of K_{ATP}-HI—diffuse hyperinsulinism (DiCHI) and focal hyperinsulinism (FoCHI). Cases of FoCHI account for approximately 40–60% of all cases of K_{ATP}-HI in which pancreatectomy is required. In FoCHI, a somatic reduction to homozygosity (or hemizygosity) of a paternally inherited mutation of the *ABCC8* or *KCNJ11* gene and a specific loss of maternal alleles of the imprinted chromosome region 11p15 result in a focal lesion composed of hyperplastic islet-cell clusters of clonal origin (focal adenomatosis).[17] This somatic loss alters the expression of imprinted genes of region 11p15.5, including genes involved in cell proliferation such as *H19*,

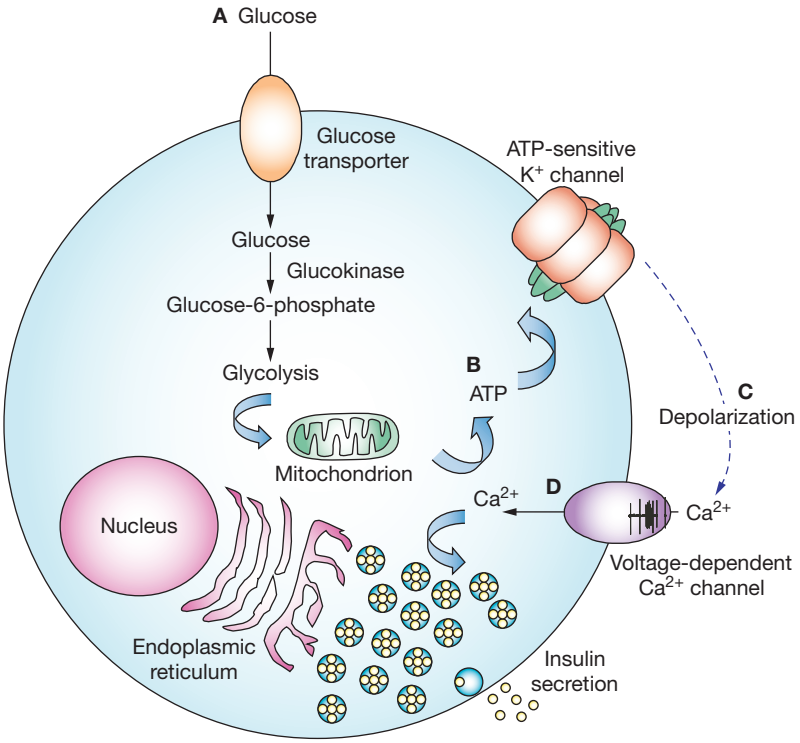

Figure 1 Glucose-stimulated insulin secretion in pancreatic β-cells. In the unstimulated state, the β-cell ATP-sensitive potassium channels are open, keeping a resting membrane potential of approximately –65 mV. (**A**) Following the uptake of glucose and its metabolism by glucokinase, (**B**) an increase in the intracellular ATP:ADP ratio results in closure of ATP-sensitive potassium channels, (**C**) depolarization of the cell membrane and subsequent opening of voltage-dependent Ca^{2+} channels. (**D**) The resulting increase in cytosolic Ca^{2+} concentration triggers insulin release.

CDKN1C (cyclin-dependent kinase inhibitor 1 C; formerly *p57^{KIP2}*), and the gene encoding insulin-like growth factor 2 (*IGF2*). *H19* (which encodes untranslated messenger RNA) and *CDKN1C*, expressed on the maternal allele, are antiproliferative, tumor-suppressor genes, whereas *IGF2* is expressed on the paternal allele and is a growth-promoting gene.

H19 and *CDKN1C* expression is significantly reduced in focal lesions.[18] Although expression of *IGF2* in these lesions is not consistently increased, the imbalance between *IGF2* and *H19* expression is thought to have an important role in the islet adenomatosis.[19] The somatic event of loss of heterozygosity probably occurs during embryonic life and does not repeat, since the presentation is at birth and no recurrent cases after removal of the lesion have been reported. Notably, loss of heterozygosity of the same 11p15 region has been found in some insulinomas.[18]

In DiCHI, β-cells throughout the pancreas are functionally abnormal and have characteristic enlarged nuclei in about 2–5% of cells (Figure 2A). FoCHI lesions are usually less than 10 mm in diameter and are characterized by the presence of a confluent proliferation of endocrine cells (adenomatous hyperplasia). Some of the β-cells within the focal lesion contain enlarged nuclei, but the absence of abnormal or enlarged islet cell nuclei in nonadjacent pancreas to the focal lesions is key to the classification as focal rather than diffuse hyperinsulinism.

In contrast to true adenomas, the focal adenomatous hyperplasia includes exocrine acinar cells intermixed within the lesion (Figure 2B). The morphology of islets away from the focal lesion is normal.[20] It has been suggested that islets adjacent to the lesions exhibit a resting appearance.[18] In the past, the term nesidioblastosis, which describes the persistence of diffuse proliferation of islet cells budding from pancreatic ducts, was believed to be of pathologic significance in children with CHI.[21] Although the term continues to appear in the literature,[21] it has been recognized that nesidioblastosis is a normal feature of the pancreas in early infancy[22] and should not be used to refer to lesions associated with hyperinsulinism.

Glutamate dehydrogenase hyperinsulinism

Pathophysiology and genetics

The second-most-common form of CHI, GDH-HI, also known as the hyperinsulinism and hyperammonemia syndrome (reviewed by Stanley[23]), is caused by gain-of-function mutations of a mitochondrial enzyme, GDH.[8] This enzyme is a key regulator of amino acid and ammonia metabolism in pancreatic β-cells, liver, and brain. GDH is normally activated by leucine and ADP, with GTP and ATP acting as allosteric inhibitors. In pancreatic β-cells, leucine stimulates insulin secretion by allosterically activating GDH to increase oxidation of glutamate to α-ketoglutarate, thereby raising the ATP:ADP ratio and triggering insulin release in a K_{ATP}-channel-dependent way (Figure 3).

In GDH-HI, missense mutations of GDH reduce the sensitivity of the enzyme to allosteric inhibition by GTP. The loss of inhibitory control of GDH in pancreatic β-cells results in excessive flux through GDH and excessive insulin release. Isolated islets from transgenic mice expressing a mutated human GDH exhibit normal glucose-stimulated insulin secretion

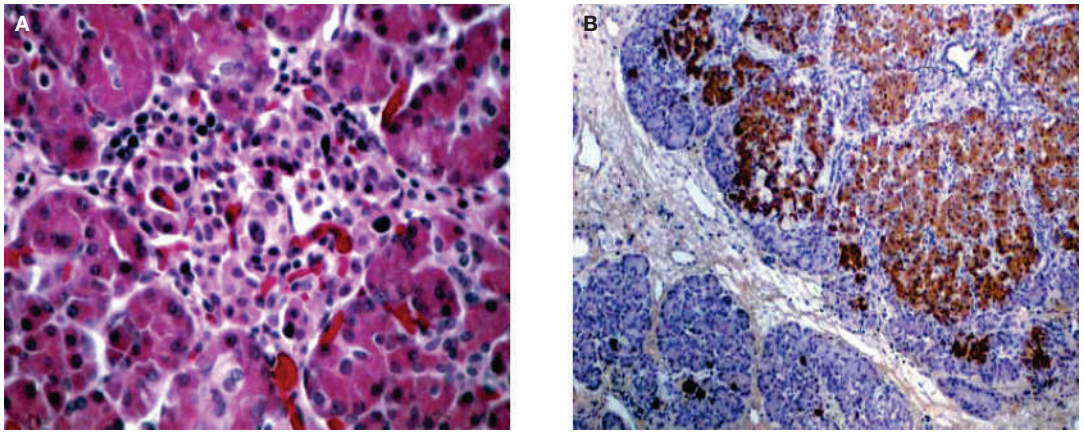

Figure 2 Histological forms of congenital hyperinsulinism. (**A**) Pancreatic section from a patient with diffuse hyperinsulinism. Note the enlarged and hyperchromatic nuclei in an otherwise normal-appearing islet (hematoxylin and eosin stain; original magnification, ×400). (**B**) Focal adenomatosis (focal hyperinsulinism). Note the normal-appearing pancreas surrounding the focal hyperplasia (insulin immunostaining shows brown, with hematoxylin counterstain; original magnification, ×40).

but enhanced leucine-stimulated and amino-acid-stimulated insulin secretion.[24] In the liver, increased GDH activity leads to excessive ammonia production and impaired urea synthesis (Figure 3). The consequences of increased enzyme activity in the brain are less clear, but might explain the lack of toxic effects of hyperammonemia in affected children. *De novo* (80%) and dominantly inherited (20%) mutations have been reported in the GTP-inhibitory allosteric binding site or in an antenna region of the enzyme, which has a role in communicating with adjacent enzyme subunits.

Phenotype

The hallmark of GDH-HI is recurrent episodes of fasting and postprandial hypoglycemia associated with persistent asymptomatic elevation of plasma ammonia levels. In GDH-HI, the hypoglycemia is less severe than in K_{ATP}-HI and can be precipitated by a protein meal.[25] Cases are frequently not diagnosed until several months of age, given the lack of severe symptoms at birth; furthermore, these infants are usually not large for gestational age, as seen in K_{ATP}-HI. Plasma ammonia levels are mildly elevated (two to five times the upper limit of normal), and stable with fasting and protein meals. The hyperammonemia seems to be asymptomatic. Children with GDH-HI can present with an unusual pattern of generalized seizures.[26] The fasting and postprandial hypoglycemia in patients with GDH-HI is easily controlled with diazoxide.

Glucokinase congenital hyperinsulinism

A less frequent form of CHI, glucokinase-HI (GK-HI), is caused by activating mutations in *GCK*, which encodes glucokinase,[7] a hexokinase that serves as a glucose sensor in pancreatic β-cells and seems to have a similar role in enteroendocrine cells, hepatocytes, and hypothalamic neurons. In β-cells, glucokinase controls the rate-limiting step of glucose metabolism and is responsible for glucose-stimulated insulin secretion[27] (Figure 1). In GK-HI, activating mutations result in increased affinity of glucokinase for glucose, closure of K_{ATP} channels and inappropriate insulin secretion. The β-cell glucose threshold for glucose-stimulated insulin secretion in children with GK-HI may therefore be as low as 1.5 mmol/l (27 mg/dl), whereas the normal glucose threshold is maintained close to 5 mmol/l (90 mg/dl) in humans. Inactivating mutations in glucokinase result in maturity-onset diabetes of the young in the heterozygous state and permanent neonatal diabetes in homozygotes.[27] Both activating and inactivating mutations are inherited in an autosomal dominant manner.

GK-HI is a rare cause of CHI; five mutations have been reported to date.[28] The age of onset and severity of symptoms vary markedly.[7,29–31] Some mutations seem to have a mild phenotype with fasting hypoglycemia that is responsive to pharmacological treatment. Others seem to lower the glucose threshold further and might be more difficult to treat.[31] Similarly, islet morphology seems to vary according to the

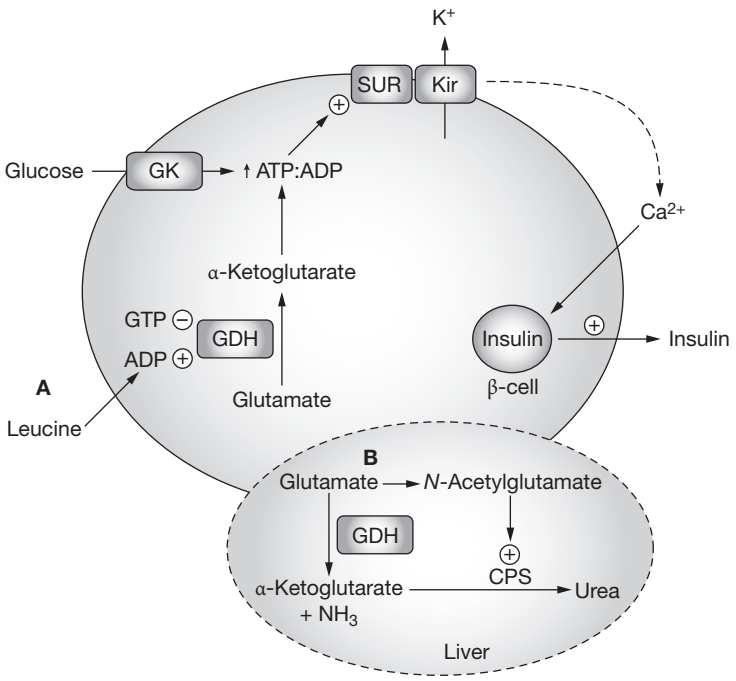

Figure 3 Mechanisms of hyperinsulinism and hyperammonemia in GDH hyperinsulinism. (**A**) In pancreatic β-cells, leucine stimulates insulin release by allosterically activating GDH to increase oxidation of glutamate, thereby raising the ATP:ADP ratio and activating the ATP-sensitive potassium channel to stimulate insulin release (see Figure 1). The loss of inhibitory control of GDH leads to excessive flux through GDH and excessive release of insulin. (**B**) In the liver, elevated activity of GDH may lead to hyperammonemia through two mechanisms: increased rates of NH_3 release from the amino nitrogen of glutamate; and limited disposal as a result of a decreased pool of hepatic glutamate causing decreased CPS activity. Abbreviations: CPS, carbamoylphosphate synthetase; GDH, glutamate dehydrogenase; GK, glucokinase; Kir, Kir6.2; SUR, sulfonylurea receptor 1.

genotype, with normal-appearing islets in some cases,[30] and enlarged islet size in others.[31]

Short-chain 3-hydroxyacyl-CoA dehydrogenase hyperinsulinism

Recently, a mutation in *HADH*, the gene encoding the mitochondrial enzyme SCHAD, was found to be associated with CHI.[9,32,33] SCHAD catalyzes the third of four steps in the mitochondrial fatty acid oxidation spiral. SCHAD-HI is an autosomal recessive disorder characterized by fasting hypoglycemia due to inappropriate insulin regulation. The biochemical hallmark, in addition to markers of increased insulin action, is increased levels of plasma 3-hydroxybutyryl-carnitine and increased levels of 3-hydroxyglutarate in urine. In contrast to all other defects in fatty acid oxidation, children with SCHAD-HI have no signs of hepatic dysfunction, cardiomyopathy, or effects on skeletal muscle.[32] The clinical presentation of SCHAD-HI is heterogeneous, ranging from late onset of mild hypoglycemia to severe early onset of hypoglycemia in the neonatal period. The hypoglycemia in patients with SCHAD-HI is responsive to medical therapy with diazoxide. Although multiple potential mechanisms have been postulated,[34] the cause of dysregulated insulin secretion in SCHAD deficiency remains to be elucidated.

OTHER FORMS OF HYPERINSULINISM IN NEONATES

Perinatal stress-induced hyperinsulinism

In addition to genetic disorders of insulin secretion, hyperinsulinism resulting in prolonged neonatal hypoglycemia can present in the neonatal period in infants exposed to perinatal stress such as birth asphyxia, maternal toxemia, prematurity, or intrauterine growth retardation. Although the first reports of this form of hyperinsulinism date back to 1984,[35] the condition has been largely ignored. Up to 10% of small for gestational age infants might have this form of hyperinsulinism (CA Stanley, unpublished observations).

The clinical presentation of perinatal stress-induced hyperinsulinism is characterized by high glucose utilization, and the response to fasting hypoglycemia shows an elevated plasma insulin level (although it might be normal in some), low β-hydroxybutyrate and fatty acid levels, and a glycemic response to glucagon. Unlike the transient hyperinsulinism seen in the infants of diabetic mothers, perinatal stress-induced hyperinsulinism can persist for several days to several weeks. In a series of neonates diagnosed after 1 week of age, the median age of resolution was 6 months.[36] The mechanism responsible for the dysregulated insulin secretion is not known. Acute insulin response (AIR) testing shows that, in general, the patterns of insulin response to secretagogues (calcium, tolbutamide, glucose and leucine) in infants with prolonged neonatal hyperinsulinism resembled those of normal controls.[36] These infants usually respond very well to medical therapy with diazoxide.

Hyperinsulinism in Beckwith–Wiedemann syndrome

Beckwith–Wiedemann syndrome (BWS) is a clinically and genetically heterogeneous disorder characterized by somatic overgrowth, physical

characteristics such as macroglossia, hemihypertrophy and transverse creases of the ear lobes, hypoglycemia, and predisposition to childhood tumors. Among cases of BWS, 85% are sporadic and 15% are autosomal dominant. Although most children with BWS are karyotypically normal, genetic abnormalities in the imprinted region of chromosome 11 with aberrant expression of multiple imprinted growth regulatory genes, including, *H19*, *IGF2*, and *CDKN1C*, has been described in BWS. Among sporadic and autosomal dominant pedigrees, mutations in *CDKN1C* have been observed in a small number of cases. In sporadic cases, the DNA alterations involving 11p15 include translocations or inversions, duplications, and uniparental disomy (for a detailed review, see Li *et al.*[37]). Hypoglycemia occurs in up to 50% of patients with BWS (reviewed by Dunne *et al.*[10]). The clinical presentation is variable—mild and transient in some, but severe and persistent in others.

At least two theories have been proposed to explain the hypoglycemia: one involves the insulin-like actions of IGF2, and the second involves dysregulated insulin secretion. The underlying mechanism of hyperinsulinism in these patients is not clear. Insulin responses to leucine and arginine have been reported normal (reviewed by Munns and Batch[38]). A recent report described loss of function of β-cell K_{ATP} channels in a patient with BWS as a result of uniparental disomy for chromosome 11p15.[39] Limited histological studies have described diffuse islet hyperplasia and hypertrophy (reviewed by Munns and Batch[38]). The heterogeneous phenotype can probably be explained by the variable genotype, and it is possible that the mechanisms of hypoglycemia are also heterogeneous. Similarly, the response to medical therapy in BWS is variable, with some cases well controlled with inhibitors of insulin secretion, whereas others require partial pancreatectomy. For reasons that are not clear, in most cases the hypoglycemia resolves spontaneously.[40]

Hyperinsulinism in congenital disorders of glycosylation

Congenital disorders of glycosylation (CDG) are inherited metabolic diseases caused by defects in the biosynthesis or transfer of lipid-linked oligosaccharides to the nascent protein chain (type I) or compromised processing of protein-bound oligosaccharides (type II). In CDG, hypoglycosylation of different glycoproteins leads to a variety of symptoms affecting multiple systems, such as the brain, liver, gastrointestinal system, and skeleton, among others.[41] Hypoglycemia with features of hyperinsulinism has been reported in cases of CDG-Ia, caused by phosphomannomutase deficiency;[42] CDG-Ib, caused by phosphomannose isomerase deficiency;[43,44] and more recently in a case of CDG-Id, caused by dolichyl-P (Dol-P)-mannose (Man):Man_5-*N*-acetyl glucosamine $(GlcNAc)_2$-PP-Dol-α-1,3-mannosyltransferase deficiency.[45] The pathophysiology behind the dysregulated insulin secretion in these conditions is unknown; hypoglycosylation of the sulfonylurea receptor or other proteins involved in insulin secretion has been speculated to be the responsible mechanism, whereas Sun *et al.*[45] reported islet cell hyperplasia in a patient with CDG-Id and hyperinsulinism. Some of the described patients were successfully treated with diazoxide.

DIAGNOSTIC APPROACH TO TREAT HYPERINSULINISM

Diagnosis of hyperinsulinism

The diagnosis of hyperinsulinism is based on the clinical presentation and the biochemical markers at the time of hypoglycemia. The index of suspicion should be high in large or small for gestational age infants and infants exposed to perinatal stress, as well as infants from families with a history of CHI.

Infants with CHI present with severe and persistent hypoglycemia manifested by seizures, lethargy, apnea and other symptoms resulting from neuroglucopenia, and increased glucose requirements (greater than 10 mg/kg/min). Plasma insulin levels are rarely dramatically elevated in CHI; rather, there is inadequate suppression of insulin at low plasma glucose concentrations. The diagnosis of hyperinsulinism in infants is thus most frequently based on evidence of excessive insulin action, such as suppression of plasma β-hydroxybutyrate and free fatty acid levels. Similarly, a glycemic response greater than 1.7 mmol/l (30 mg/dl) to glucagon at the time of hypoglycemia indicates excess insulin action and is useful for confirming the diagnosis (Box 2). Additional tests for specific forms of CHI include plasma ammonia levels (GDH-HI), plasma acyl-carnitine profile (elevated 3-hydroxybutyryl-carnitine) and urine organic acids (3-hydroxyglutarate in urine; SCHAD-HI).

Genetic testing is available for four of the five genes known to be associated with CHI through

Box 2 Diagnostic congenital hyperinsulinism criteria based on the 'critical blood sample'.

- Hyperinsulinemia (plasma insulin >2 μU/ml, depending on sensitivity of insulin assay); note that the absence of hyperinsulinemia does not rule out congenital hyperinsulinism
- Hypofattyacidemia (plasma free fatty acids <1.5 mmol/l)
- Hypoketonemia (plasma β-hydroxybutyrate <2.0 mmol/l)
- Glycemic response to 1 mg intravenous glucagon (Δ glucose >30 mg/dl)

commercial laboratories. In addition, AIR tests are useful in phenotypic characterization: patients with diffuse K_{ATP}-HI display abnormal positive responses to calcium, abnormal negative response to the K_{ATP}-channel antagonist, tolbutamide, and impaired responses to glucose.[46,47] By contrast, infants with GDH-HI exhibit increased responses to leucine.[48] The response to leucine is thus used to differentiate between K_{ATP}-HI and GDH-HI.

Diagnosis of diffuse versus focal congenital hyperinsulinism

In recent years, efforts have been made to discriminate between DiCHI and FoCHI, given the potential for surgical cure of FoCHI. The clinical presentation of DiCHi and FoCHi is indistinguishable. Similarly, AIR tests cannot reliable differentiate between DiCHI and FoCHI.[49,50]

Interventional radiology studies, such as transhepatic portal venous insulin sampling[51] and selective pancreatic arterial calcium stimulation,[49] have been used to localize focal lesions. Both have only modest success and are technically difficult and highly invasive. More recently, PET scans with flurorine-18 L-3,4-dihydroxyphenylalanine (^{18}F-fluoro-L-DOPA) have been shown to accurately discriminate FoCHI from DiCHI.[52–54] It has been previously shown that β-cells take up L-DOPA[55] and that DOPA decarboxylase is active in pancreatic islet cells.[56] In children with FoCHI there is local accumulation of ^{18}F-fluoro-L-DOPA, and coregistration of PET and MRI images allows the anatomical localization of the lesion. Diffuse pancreatic accumulation of ^{18}F-fluoro-L-DOPA is consistent with DiCHI; furthermore, ^{18}F-fluoro-L-DOPA PET scan findings have been confirmed by immunohistochemical detection of DOPA decarboxylase in the pancreatic specimens, and administration of a DOPA decarboxylase inhibitor abolishes the uptake.[57]

MANAGEMENT OF HYPERINSULINISM

The management of infants with hypoglycemia should be guided by the following goals: to prevent brain damage from recurrent hypoglycemia; to establish a specific diagnosis and therapy; and to encourage normal feeding behavior while assuring safe fasting tolerance. The new information about the molecular causes of CHI makes specific diagnoses and disease-specific treatment feasible. A multidisciplinary approach is key to the management of these children (Figure 4). The first step in management is correction of hypoglycemia with the therapeutic goal of maintaining plasma glucose levels above 3.9 mmol/l (70 mg/dl). Since infants with CHI can require glucose infusion rates as high as 20–30 mg/kg/min, appropriate venous access should be established.

Medical therapy

Once the diagnosis of CHI has been made, medical therapy with diazoxide should be started. Diazoxide is a K_{ATP}-channel inhibitor and therefore requires functional channels to be present at the cell surface to have an effect; thus, most patients with K_{ATP}-HI do not respond to diazoxide. Patients with GDH-HI, GK-HI, SCHAD-HI and those with perinatal stress-induced hyperinsulinism respond to diazoxide. The usual dose of diazoxide is 5–15 mg/kg/day, given orally once or twice a day. The major adverse event with diazoxide in neonates is fluid retention. Concomitant use of a diuretic (chlorothiazide or furosemide) should be considered, especially in infants receiving intravenous fluids.

The half-life of diazoxide is 24–36 h in adults; limited data indicate that in children the half-life is 9.5–24.0 h.[58] The response to diazoxide should thus be evaluated after at least 5 days of therapy. Successful response should be demonstrated by showing maintenance of plasma glucose above 3.9 mmol/l (70 mg/dl) after fasting. The duration of the fast should be determined on a case-by-case basis, based in part on the age of the patient.

The second line of medical therapy for infants unresponsive to diazoxide is octreotide. This long-acting somatostatin analog inhibits insulin secretion by inducing hyperpolarization of β-cells, direct inhibition of voltage-dependent calcium channels, and more distal events in the

insulin secretory pathway. Octreotide is administered subcutaneously every 6–8 h at 5–20 μg/kg/day, or as a continuous infusion. The initial response to octreotide is good in most infants with CHI, but tachyphylaxis develops after a few doses, rendering therapy inadequate for long-term use in most cases.

The use of calcium-channel blockers to decrease voltage-dependent calcium-channel activity has been proposed as an alternative medical therapy for diazoxide-unresponsive cases. A few reports of successful treatment with nifedipine are published (reviewed by Müller *et al.*[59]), but there is no long-term experience with nifedipine and most centers have not had success with this drug.

Glucagon, given as a continuous intravenous infusion (1 mg/day) can help maintain euglycemia in infants waiting for surgery (Box 3). A nonsurgical approach in the management of infants with medically unresponsive CHI, consisting of continuous glucagon and octreotide infusion in combination with continuous enteral feedings, has been proposed.[60] The long-term outcome of this approach has not been evaluated.

Surgical therapy

Surgical therapy, particularly for focal K_{ATP}-HI, requires a multidisciplinary approach involving pediatric endocrinologists, interventional radiologists, pathologists, and surgeons with expertise in this disorder. When medical therapy fails and surgery is being contemplated, transfer to a specialized center should be considered (Figure 4).

A systematic approach directed to the differentiation of focal versus diffuse disease and the anatomic localization of focal lesions should be undertaken before surgery. In addition to the ^{18}F-fluoro-L-DOPA PET scan, intraoperative frozen section evaluation has proven to have an essential role in identifying focal lesions and guiding the extent of the surgery.[61] This management approach can prevent the need for a near-total pancreatectomy and its related complications in subjects with focal disease. Although limited to a few cases, the use of laparoscopy to localize and enucleate focal lesions is a novel and promising approach.[62] Infants with diffuse disease will normally require a near-total pancreatectomy (95–98%) to control the hyperinsulinism and might require additional therapy with diazoxide, octreotide, and/or frequent feedings to maintain euglycemia. Some of these patients require additional resection because of the severity of the hypoglycemia.

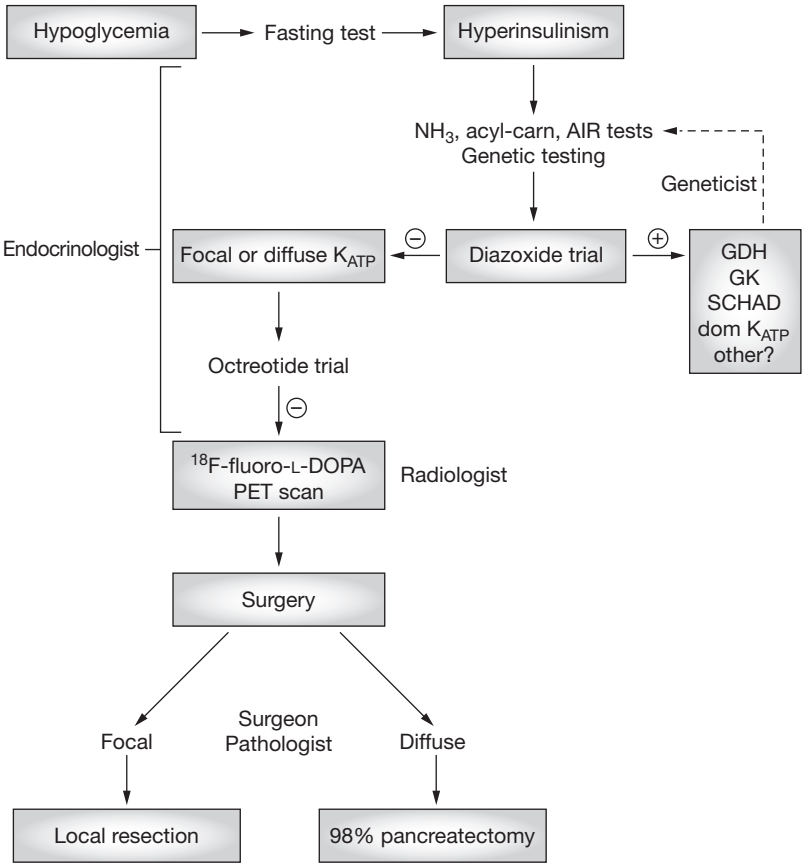

Figure 4 Current management paradigm for congenital hyperinsulinism. Once the diagnosis of hyperinsulinism has been established through the evaluation of the 'critical blood sample' (see Box 2), a treatment trial with diazoxide should be started. Additional testing, such as NH_3 levels, acyl-carn profile, AIR testing and genotyping, helps to make the diagnosis of the specific forms of congenital hyperinsulinism. In most cases, failure to respond to diazoxide indicates K_{ATP} hyperinsulinism and a trial of octreotide should be added to the treatment regimen. In children unresponsive to medical therapy and thus requiring surgery, efforts should be directed to distinguish and localize focal lesions that can be focally resected. This approach requires a multidisciplinary team comprising neonatologists, endocrinologists, radiologists, pathologists, geneticists, and surgeons. Abbreviations: ^{18}F-fluoro-L-DOPA, flurorine-18 L-3,4-dihydroxyphenylalanine; acyl-carn, acyl-carnitine profile; AIR, acute insulin response; dom K_{ATP}, autosomal dominant mutations in ATP-dependent potassium channel; GK, glucokinase; GDH, glutamate dehydrogenase; SCHAD, short-chain 3-hydroxyacyl-CoA dehydrogenase.

PROGNOSIS AND OUTCOME

Limited data on the long-term outcome in children with CHI are available, but these children are at risk of severe complications inherent to their disease and as a consequence of their therapy. The risk of developing diabetes mellitus has been attributed to pancreatectomy,[63] but it is clear than even patients who did not undergo surgical therapy can develop diabetes mellitus later in life (CA Stanley, unpublished observations).

Box 3 Treatment of congenital hyperinsulinism.

Intravenous glucose infusion
Dextrose emergency bolus: 0.2 g/kg (2 ml/kg of 10% dextrose)
Maintenance glucose infusion: 8–10 mg/kg/min, increase infusion as necessary to maintain blood glucose >70 mg/dl

Diazoxide
5–15 mg/kg/day orally divided into two doses; side effects include fluid retention and hypertrichosis

Octreotide
5–20 μg/kg/day subcutaneously divided in three or four doses; side effects include diarrhea, abdominal discomfort and gallstones

Glucagon
1 mg/day given as a continuous intravenous infusion; side effects include vomiting and rebound hypoglycemia after discontinuation

KEY POINTS

- Hyperinsulinism is the most common cause of persistent hypoglycemia in infants and children
- To date, mutations in five genes are known to encode proteins that cause congenital hyperinsulinism: sulfonylurea receptor 1 and Kir6.2 (the two components of the ATP-dependent potassium channel), glutamate dehydrogenase, glucokinase, and short chain 3-hydroxyacyl-CoA dehydrogenase
- ATP-dependent potassium channel hyperinsulinism is the most common and severe form of hyperinsulinism and can present as diffuse or focal disease; this disease can be cured by surgical resection
- A multidisciplinary approach is important in the management of infants with congenital hyperinsulinism; the goal of therapy is to avoid neurodevelopmental complications by preventing and treating hypoglycemia

In a large series of 114 patients with CHI, the incidence of diabetes mellitus was as high as 27% after pancreatectomy and the highest rate (71%) was seen in patients who had undergone more than one surgical resection. In the same series, a high frequency (44%) of neurodevelopmental retardation was reported.[64] In another series of 90 patients with CHI, severe mental retardation was found in 8%, with less severe disability in 18%. These authors found that psychomotor retardation is more common in patients with neonatal hypoglycemia than in those with onset of hypoglycemia during infancy.[65] In our experience, the prevalence of developmental delay in patients with CHI is around 30%; patients with CHI requiring surgical therapy have a higher incidence of neurodevelopmental problems than patients responsive to medical therapy.[66]

CONCLUSIONS

The identification of the molecular causes of CHI has allowed us to understand the clinical heterogeneity of patients with this condition. Similarly, the recognition of the two different histological presentations of K_{ATP}-HI has resulted in the development of management protocols aimed to distinguish between diffuse and focal disease and to localize focal lesions to guarantee surgical cure. As little advancement in the development of effective medical therapy has been made in the past, efforts should be directed to this end in the next few years.

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