Evidence for a Continuum of Genetic, Phenotypic, and Biochemical Abnormalities in Children with Growth Hormone Insensitivity

Alessia David, Vivian Hwa, Louise A. Metherell, Irène Netchine, Cecilia Camacho-Hübner, Adrian J. L. Clark, Ron G. Rosenfeld, and Martin O. Savage

Department of Endocrinology (A.D., L.A.M., A.J.L.C., M.O.S.), William Harvey Research Institute, Barts and the London School of Medicine and Dentistry, London EC1M 6BQ, United Kingdom; Department of Pediatrics (V.H., R.G.R.), Oregon Health and Science University, Portland, Oregon 97239; Hôpital Armand-Trousseau (I.N.), Explorations Fonctionnelles Endocriniennes, 75012 Paris, France; Université Pierre et Marie Curie (I.N.), 75005 Paris, France; Institut National de la Santé et de la Recherche Médicale (I.N.), Unité 938, Team 4, 75012 Paris, France; and Karolinska Institute (C.C.-H.), 171 65 Stockholm, Sweden

GH insensitivity (GHI) presents in childhood as growth failure and in its severe form is associated with dysmorphic and metabolic abnormalities. GHI may be caused by genetic defects in the GH–IGF-I axis or by acquired states such as chronic illness. This article discusses the former category. The field of GHI due to mutations affecting GH action has evolved considerably since the original description of the extreme phenotype related to homozygous GH receptor (*GHR*) mutations over 40 yr ago. A continuum of genetic, phenotypic, and biochemical abnormalities can be defined associated with clinically relevant defects in linear growth. The role and mechanisms of the GH–IGF-I axis in normal human growth is discussed, followed by descriptions of mutations in *GHR*, *STAT5B*, *PTPN11*, *IGF1*, *IGFALS*, *IGF1R*, and *GH1* defects causing bioinactive GH or anti-GH antibodies. These defects are associated with a range of genetic, clinical, and hormonal characteristics. Genetic abnormalities causing growth failure that is less severe than the extreme phenotype are emphasized, together with an analysis of height and serum IGF-I across the spectrum of different types of *GHR* defects. An overall view of genotype and phenotype relationships is presented, together with an updated approach to the assessment of the patient with GHI, focusing on investigation of the GH–IGF-I axis and relevant molecular studies contributing to this diagnosis. **(*Endocrine Reviews* 32: 472–497, 2011)**

ISSN Print 0021-972X ISSN Online 1945-7197
Printed in U.S.A.

doi: 10.1210/er.2010-0023 Received October 22, 2010. Accepted March 16, 2011.
First Published Online April 27, 2011

Abbreviations: ALS, Acid-labile subunit; GHBP, GH binding protein; GHI, GH insensitivity; GHR, GH receptor; GHRE, GH-responsive element; IGFBP, IGF-binding protein; IGFGT, IGF-I generation test; IGFIR, IGF-I receptor; IGHD, isolated GH deficiency; IUGR, intrauterine growth retardation; JAK2, Janus kinase 2; NS, Noonan syndrome; PI3K, phosphoinositide-3 kinase; PTPN11, protein tyrosine phosphatase, nonreceptor type 11; RAS, rat sarcoma viral oncogene homolog; rh, recombinant human; SDS, SD score; SH2, src-homology 2; SHP-2, SH2 domain phosphatase-2; STAT, signal transducer and activator of transcription.

I. Introduction

GH insensitivity (GHI) (OMIM no. 262500 and 245590) was first described in a pediatric setting by Laron *et al.* in 1966, with the description of three children with extreme growth failure from a consanguineous Jewish family of Yemenite origin who had the phenotype of hypopituitarism with high serum GH concentrations (1, 2). Although an abnormal GH molecule was initially suspected, this disorder, which for many years was referred to as "Laron syndrome," was shown to be caused by a defect in the GH receptor (GHR) resulting in the absence of binding of ^{125}I-GH to GHR prepared from the patients' liver membranes (3). This striking but very rare phenotype, which was also untreatable at that time, became synonymous with the diagnosis of GHI, a perception that remained unchallenged for over 20 yr. In the late 1980s, two pivotal developments brought important changes to the field. The first was the synthesis and availability of recombinant human (rh) IGF-I for therapy (4, 5), and the second was the advent of molecular techniques, which led to the cloning and characterization of the human *GHR*, thus initiating the understanding of the pathophysiology of GHI (6, 7). The subsequent study of genetic abnormalities in the GH–IGF axis has provided invaluable information on the physiology of human linear growth.

As may occur in medicine, scientific advances frequently move more rapidly than the perceptions of clinicians, who may remain attached to the recognized and trusted view of a certain disorder, particularly if it is rare. This observation can be made in relation to GHI, which is now known not to be a single entity, but rather to be a broad diagnostic category comprising a range of molecular defects in the GH–IGF axis. These defects, which may involve genes coding for proteins that regulate GH binding or signal transduction and IGF-I synthesis, transport, or action, are associated with an equally varied range of phenotypes and biochemical abnormalities. This review will present and discuss evidence for a continuum of GH–IGF axis abnormalities associated with a range of phenotypes consistent with GHI.

Previous reviews have described the characteristics of the extreme phenotype (2, 8, 9), so we concentrate on defects illustrating the range of phenotypes. This article discusses the genetic causes of GHI, also referred to as "primary IGF deficiency," rather than acquired states of GHI related, for example, to chronic illness or malnutrition. We aim also to update and orient clinicians in the appropriate diagnostic approach to children with growth failure. We challenge and hopefully lay to rest the view that GHI is an excessively rare disorder associated with a single historical phenotype.

II. The GH–IGF-I Axis in Human Growth

A. Physiology of GH and the IGF-I system in relation to linear growth

The actions of GH are mediated by a combination of components of the IGF system, including IGF-I, IGF-binding proteins (IGFBP), the IGF-I receptor (IGFIR), and IGF-independent effects through direct GH action. The GH–IGF-I axis is shown in Fig. 1 and discussed further in *Sections II.A and II.B*. The original "somatomedin hypothesis" proposed that GH binding to its receptor stimulated IGF-I production, which independently affected growth (10). However, the discovery that IGFs are expressed in most tissues questioned this original hypothesis. Green *et al.* (11) proposed the "dual effector hypothesis" in 1985, based on studies with adipocytes suggesting that GH regulates the expression of locally produced IGF-I, which then acts in an autocrine/paracrine manner. Expression of the *IGF1* gene was found in multiple tissues throughout embryonic and postnatal development (12, 13). In addition, injection of GH into hypophysectomized rats increased *IGF1* mRNA in numerous nonhepatic tissues (14, 15). Direct injection of GH into the cartilage growth plate of hypophysectomized rats also resulted in significantly increased longitudinal bone growth (16). These and other studies suggested that GH has local effects, independent of those mediated by the increase in circulating "endocrine" IGF-I. This hypothesis was then extended by Isaksson and colleagues (17, 18), who demonstrated that GH stimulated differentiation of preadipocytes and chondrocytes in the growth plate, whereas IGF-I stimulated their clonal expansion.

In a comprehensive review of the evolution of our understanding of the interrelated roles of GH and IGF-I, LeRoith *et al.* (19) presented a revised somatomedin hypothesis in 2001, taking account of gene-deletion experiments in mice that questioned the role of liver IGF-I and its circulating endocrine form in controlling postnatal growth and development (20, 21). Liver-specific *Igf1* knockout mice continued to grow normally despite a significant reduction in circulating IGF-I, indicating that locally produced IGF-I was an important growth mediator (19). However, LeRoith and colleagues (22) recently modified the conclusions on the role of hepatic IGF-I after data showing that when the hepatic *Igf1* transgene was inserted into the IGF-I null mouse, it elevated serum IGF-I levels and largely restored growth. In 2007, Kaplan and Cohen (23) proposed a further modified somatomedin-IGF hypothesis that considered the apparent paradox that GH exerts its effects through IGF, some of which oppose the known actions of GH. The so-called "augmentative/counteractive hypothesis" proposed that the IGF are augmentative hormones that amplify the anabolic actions of GH,

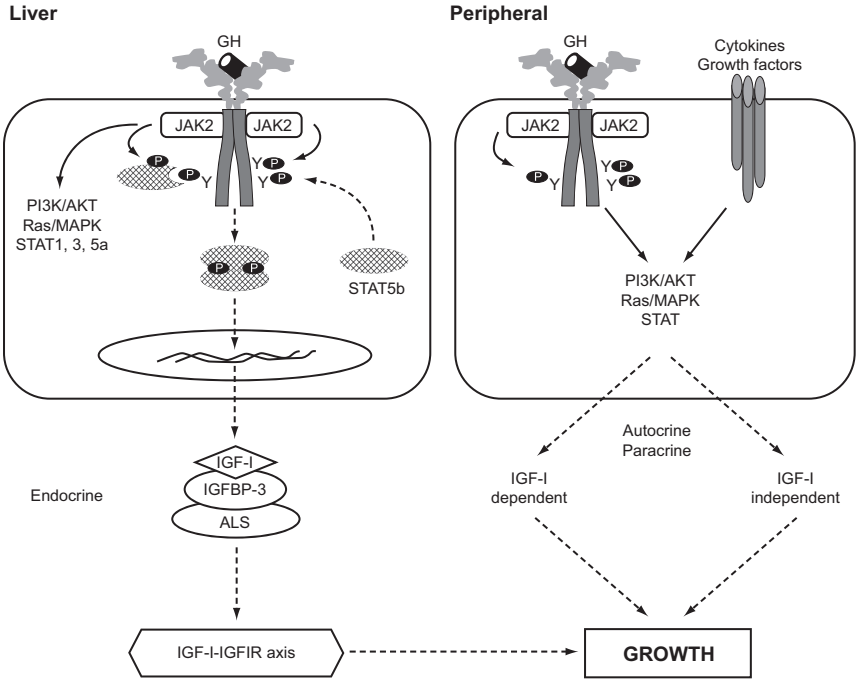

FIG. 1. The GH–IGF-I axis in human growth. See *Section II.B* for explanation. *Solid arrows*, Activation processes; *dashed arrows*, translocation processes. AKT, v-akt murine thymoma viral oncogene homolog, also known as PKB, or protein kinase B; P, phosphorylated residue; Y, tyrosine.

while countering the potentially undesirable GH effects of gluconeogenesis and lipolysis (23).

Studies using various *Igf1* knockout mouse models have shown that GH stimulates longitudinal bone growth by both IGF-I-independent and IGF-I-dependent mechanisms. About 75% of serum IGF-I is liver-derived, whereas the remainder originates from nonhepatic tissues (20, 21). In addition, serum levels of the acid-labile subunit (ALS) and IGFBP-3 are important in maintaining circulating IGF-I (24). The importance of ALS was clearly shown in the *Igfals* knockout mouse model (25) and by Domené *et al.* (26), who reported the first homozygous mutation in human *IGFALS* causing severe IGF-I deficiency.

B. Mechanisms of GH and IGF-I actions

Pituitary-derived GH exerts its growth effects primarily by regulating the expression of IGF-I in both hepatic and nonhepatic tissues (Fig. 1). In nonhepatic tissues, other growth factors and cytokines, in addition to GH, can exert local growth effects by regulating IGF-I production and/or by mechanisms independent of IGF-I expression. GH regulates IGF-I production predominantly through the signal transducer and activator of transcription (STAT)-5b signaling system. The binding of GH to the cell surface homodimeric GHR (a cytokine receptor that lacks intrinsic kinase activity) recruits and induces signal transduction through cytosolic Janus kinase 2 (JAK2). The complex signaling pathways activated include four STATs (STAT1, -3, -5a, and -5b) and the phosphoinositide-3 kinase (PI3K) and MAPK pathways. Initiation of signal transduction through the STAT5b pathway requires STAT5b to associate with one of several JAK2-phosphorylated tyrosines located on the intracellular domain of GHR (27). In contrast, these GHR intracellular tyrosines are not necessary for activation of the STAT1, STAT3, MAPK, or PI3K pathways (27, 28), but activation is likely to be still JAK2 dependent (29), although the involvement of other kinases such as Lyn has been suggested (30, 31).

STAT5b, recruited to GH-activated GHR, is subsequently phosphorylated by JAK2, whereupon the tyrosyl-phosphorlated-STAT5b forms a homodimer and translocates to the nucleus. The dimeric phosphorylated STAT5b binds to chromosomal GH-responsive elements (GHRE) and drives transcriptional regulations of STAT5b-dependent genes, including *IGF1*, *IGFBP3*, and *IGFALS*, encoding for IGF-I, IGFBP-3, and ALS, respectively. Surprisingly little is known regarding the human GHRE recognized by STAT5b. In the rat *Igf1* gene locus, seven GH-induced STAT5b response elements located at least 63 kb upstream of the *Igf1* start site or within introns were recently reported (32). It remains to be determined whether similarly located GHRE in human *IGF1* are used by STAT5b (32, 33).

IGF-I produced in the liver circulates in ternary complex with liver-derived IGFBP-3 and ALS and is delivered to IGF-I-responsive cells and tissues. The mitogenic and metabolic effects of IGF-I are mediated through type I IGFIR, a cell-surface tyrosine kinase receptor encoded by *IGF1R*. Synthesized as a single polypeptide precursor, the IGF1R undergoes posttranslational glycosylation and is proteolytically cleaved into α- and β-chains. The mature, functional receptor is a tetramer ($\alpha_2\beta_2$), with the extracellular dimeric α-subunits involved in ligand binding, whereas intrinsic tyrosine kinase activity is located within the β-subunits (34). The binding of IGF-I to IGF1R leads to receptor autophosphorylation, resulting in recruitment of cytoplasmic components of downstream signaling pathways, including the PI3K/Akt and MAPK/Erk pathways, ultimately leading to cell proliferation and other metabolic effects.

C. Effects of human GH–IGF-I axis mutations on linear growth

Normal GH secretion and the functional integrity of the IGF system are essential for normal linear growth. Defects

TABLE 1. Classification of GHI disorders with short stature

Defects of the GH–IGF-I axis
1. GHR defects
   a. Extracellular mutations
   b. Transmembrane mutations
   c. Intracellular mutations
2. GH signal transduction defects (STAT5b)
3. Mutations of SHP-2 (encoded by *PTPN11*), K-RAS, H-RAS
4. *IGF1* gene mutations or deletions
   a. Defects causing IGF-I deficiency
   b. Bioinactive IGF-I
5. ALS defects
6. IGF-I receptor (*IGFIR*) gene mutations
7. GH-neutralizing antibodies in patients with *GH* gene deletion

Acquired disorders causing GH resistance
1. Malnutrition, parenchymal liver disease, type 1 diabetes mellitus
2. Chronic inflammatory and nutritional disorders (*e.g.* juvenile chronic arthritis, Crohn's disease, celiac disease)

that have been identified to cause impaired growth are shown in Table 1. A summary of phenotypic and biochemical features in the range of GH–IGF-I axis defects is given in Table 2. Human prenatal growth is regulated principally by nutritional supplies, which influence fetal IGF-I and, perhaps, IGF-II (35). Targeted disruption of either *Igf1* or *Igf2* in mice led to a 40% reduction in fetal growth (36). The importance of normal IGF-I production in humans was confirmed by the prenatal growth failure reported in patients with *IGF1* mutations (37, 38). IGF-I action is also essential, as demonstrated by IGFIR rodent knockout studies ($Igf1r^{-/-}$), which resulted in a 55% reduction in fetal size (36), an effect also present in humans with mutations of *IGF1R* (39). Postnatal growth may be disrupted by mutations that disturb the functional integrity of the cascade of GH–GHR interaction, GH signal transduction, and IGF-I production, transport, and action (8, 9). In states of GHI resulting from impaired GH–GHR function, IGF-I deficiency is the cardinal biochemical feature. In mutations specifically involving the *IGF1* or *IGF1R*, GHR function remains intact, resulting in possible accentuation of IGF-I-independent GH effects (38). We next discuss the key molecular defects that have been identified as causes of clinically relevant impairment of linear growth.

III. Molecular Defects Causing GH Insensitivity

A. The GH receptor (GHR)

The GHR protein mediates the effects of GH on linear growth and metabolism. It is present ubiquitously, being most abundantly expressed in the liver (40). It is composed of a large extracellular domain involved in GH binding and GHR dimerization, a single transmembrane domain that anchors the receptor to the cell surface, and an intracellular domain involved in GH signaling. GH binding activates its receptor by inducing a conformational change in a preexisting GHR dimer (41, 42), which promotes the binding of JAK2 to a proline-rich box 1 region located in the proximal intracellular portion of the GHR (43) and initiates intracellular signaling.

B. *GHR* mutations associated with a range of phenotypes

Since 1966, more than 250 patients with genetic GHI have been identified worldwide (9, 44). The reported *GHR* mutations causing GHI are shown in Tables 3, 4, and 5. The most severe phenotype was described by Laron *et al.* (1, 2) in 1966 (OMIM no. 262500). Most GHI cases

TABLE 2. Summary of phenotypic and biochemical features in the range of GH–IGF-I axis defects

Gene defect/phenotype	*GHR*	*STAT5b*	*PTPN11*	*IGF1*	*IGFALS*	*IGFIR*	Bioinactive GH	*GH1* with anti-GH antibodies
Severe growth failure	+/−	+	−	+	−	−	−	+
Mild growth failure	−/+	−	+	−	+	+	+	−
Mid-face hypoplasia	+/−	+/−	−	−	−	−	−	+
Other facial dysmorphism	−	−	+	+	−	+	−	−
Deafness	−	−	−	+/−	−	−	−	−
Microcephaly	−	−	−	+	−	+	−	−
Intellectual delay	−	−	−/+	+	−	+/−	−	−
Puberty delay	+/−	+/−	+/−	−	+	−	−	−
Immune deficiency	−	+	−	−	−	−	−	−
Hypoglycemia	+	−/+	−	−	−	−	−	−
Hyperinsulinemia	−	−	−	+/−	+	−	−	−
IGF-I deficiency	+	+	−/+	+/−	+	−	+	+
IGFBP-3 deficiency	+	+	−/+	−	+	−	+	+
ALS deficiency	+	+	−/+	−	+	−	+	+
GH excess	+	+	−	+/−	+	−	−	−
GHBP deficiency	+/−	−	−	−	−	−	−	−
Homozygous or compound heterozygous mutations	+	+	−	+	+	−	−/+	+
Heterozygous mutations	−	−	+	−/+	−	+	+/−	−

+, Positive; −, negative; +/−, predominantly positive; −/+, predominantly negative.

TABLE 3. Nonsense mutations identified in the *GHR* of patients with GHI

Mutation	Exon	cDNA position and nucleotide change	Protein change (mRNA change)	Phenotype	Ethnic origin	First author, year (Ref.)
Nonsense	2	c.11G>A	p.Trp-15X	LS	Jewish-Iraqi	Shevah, 2003 (136)
	3	c.102G>A	p.Trp16X	LS	German	Pantel, 2003 (137)
	4	c.168C>A	p.Cys38X	LS	Mediterranean	Amselem, 1991 (138); Amselem, 1993 (139)
	4	c.181C>T	p.Arg43X	LS	Ecuadorian, Greek-Anatolian	Amselem, 1991 (138); Berg, 1993 (140); Putzolu, 1997 (141)
	4	c.247C>T	p.Gln65X	LS	Indian	Sobrier, 1997 (142)
	5	c.293G>A	p.Trp80X	LS	German	Sobrier, 1997 (142)
	5	c.303C>A[a]	p.Cys83X	LS	Not reported	Tiulpakov, 2005 (143)
	5	c.338dupA	p.Tyr95X	LS	Brazilian	Diniz, 2008 (44)
	5	c.420T>A	p.Cys122X	ISS	Not reported	Goddard, 1995 (144)
	6	c.476T>A	p.Leu141X	LS	Italian	Shevah, 2003 (136)
	6	c.525G>A	p.Trp157X	LS	Turkish	Sobrier, 1997 (142)
	6	c.592G>T	p.Glu180X	LS	Italian, Inuit	Fassone, 2007 (145); Fang, 2008 (146)
	6	c.601G>T	p.Glu183X	LS	Ecuadorian, Jewish	Berg, 1994 (147)
	7	c.703C>T	p.Arg217X	LS	Jewish-Yemenite, Palestinian-Arab	Amselem, 1993 (139); Berg, 1993 (140)
	7	c.724G>T	p.Glu224X	LS	Not reported	Kaji, 1997 (148)

Protein nomenclature is based on mature GHR peptide numbering, which does not include the 18 amino acid residues of the signal peptide. c, Coding DNA sequence where nucleotide 1 is the A of the ATG-translation initiation codon and the transcript includes exon 3; ISS, idiopathic short stature; LS, Laron syndrome; p, protein sequence; X, stop codon.

[a] This mutation was reported using the old GHR numbering as 346C>A.

have autosomal recessive inheritance, and in the vast majority, a molecular defect has been identified involving either homozygous or compound heterozygous mutations (44). Because previous reviews have focused on those mutations that cause the most severe GHI phenotypes (2, 8, 9), we will emphasize *GHR* mutations causing milder phenotypes that support the concept of a continuum.

More than 70 mutations of *GHR* have been identified to date, ranging from deletions to point mutations including missense, nonsense, and splice mutations (9, 44, 45). Splice mutations represent approximately 20% of *GHR* defects and usually disrupt major regulatory elements such as the donor and acceptor splice sites. Among the defects causing aberrant *GHR* splicing, an intronic base change leading to the activation of a pseudoexon sequence and insertion of 36 new amino acids within the receptor extracellular domain was first reported in a consanguineous Pakistani family with mild GHI (46). This mutation leads to recognition of the pseudoexon and inclusion of an additional 108 bases between exons 6 and 7, leading to impaired function of the mutant protein (47). Intronic mutations resulting in pseudoexon activation are rare in genetic diseases (48). The phenotypes (see *Section IV.A*) occurring with this mutation range from severe to mild growth failure (49). A mild phenotype of GHI is also associated with heterozygous *GHR* mutations causing a dominant negative effect (50, 51). These splice site mutations (*c.876−1G>C*) and (*c.945+1G>A*) form heterodimers with the wild-type GHR and exert a dominant negative effect on the normal protein.

Differing phenotypes within the same family may also occur, as reported with one sibling having extreme growth failure [adult height, −8.7 SD score (SDS)] and a second sibling having a milder phenotype (adult height, −6.0 SDS) (28). Both siblings had the same homozygous 22-bp deletion in the cytoplasmic domain of the GHR, resulting in a frameshift and premature stop codon. The resultant GHR was truncated at amino acid 449 (GHR1–449) after box 1, the JAK2 binding domain of the receptor, and functional studies in HEK293 and Chinese hamster ovary cells showed a selective loss of STAT5 signaling in cells expressing GHR1–449 (28).

A mild phenotype was also reported in two patients with compound heterozygous mutations (52). Both had undisputed GHI, but functional studies suggested incomplete GHR defects that determined the phenotype by an additive effect of each heterozygous mutation. Recently, a further case of mild GHI was reported in a child with a height of −4.0 SDS associated with intracellular compound heterozygous mutations consisting of a *p.R211H* mutation and a novel duplication of a nucleotide in exon 9 (*c.899dupC*), the latter resulting in a frameshift and a premature stop codon (53). A recent report of a child and his mother with short stature and elevated GH binding protein (GHBP) levels associated with a novel heterozygous C→A transversion at position c.785-3 at the accep-

TABLE 4. Missense mutations identified in the *GHR* of patients with GHI

Mutation	Exon	cDNA position and nucleotide change	Protein change (mRNA change)	Phenotype	Ethnic origin	First author, year (Ref.)
Missense	2	c.1A>T	p.Met-18Leu	LS	Not reported	Quinteiro, 2002 (149)
	4	c.166T>A	p.Cys38Ser	LS	Algerian	Sobrier, 1997 (142)
	4	c.173C>T	p.Ser40Leu	LS	Turkish	Sobrier, 1997 (142)
	4	c.178G>A	p.Glu42Lys	LS	Chinese	Chen, 2003 (150)
	4	c.184G>A	p.Glu44Lys	ISS	Not reported	Goddard, 1995 (144)
	4	c.193T>C	p.Ser47Pro[a]	LS	Chinese	Ying, 2007 (151)
	4	c.202T>C	p.Trp50Arg	LS	German	Sobrier, 1997 (142)
	4	c.266G>A	p.Arg71Lys	LS	Mediterranean	Amselem, 1993 (139)
	5	c.310T>G	p.Tyr86Asp	LS	Iranian	Shevah, 2004 (152)
	5	c.335G>C	p.Cys94Ser	LS	Austrian-Caucasian	Fang, 2007 (52)
	5	c.341T>C	p.Phe96Ser	LS	Mediterranean	Amselem, 1989 (7)
	5	c.364T>G	p.Trp104Arg	LS	Turkish	Arman, 2010 (153)
	5	c.428T>C	p.Val125Ala	LS	European	Amselem, 1993 (139)
	6	c.446C>A	p.Pro131Gln	LS	Vietnamese	Walker, 1998 (154)
	6	c.485T>A	p.Val144Asp	LS	Mediterranean	Amselem, 1993 (139)
	6	c.485G>A	p.Val144Ile	ISS	Cuban (White)	Sanchez, 1998 (155)
	6	c.504T>G	p.His150Gln	LS	Austrian-Caucasian	Fang, 2007 (52)
	6	c.508G>C	p.Asp152His	LS	Indian, Pakistani	Duquesnoy, 1994 (156)
	6	c.509A>G	p.Asp152Gly	LS	Taiwanese	Yang, 2004 (157)
	6	c.512T>C	p.Ile153Thr	LS	Caucasian	Wojcik, 1998 (158)
	6	c.515A>C	p.Gln154Pro	LS	Hispanic	Wojcik, 1998 (158)
	6	c.518T>G	p.Val155Gly	LS	Saudi Arabian	Wojcik, 1998 (158)
	6	c.535C>T	p.Arg161Cys	LS, ISS	Middle-Eastern	Amselem, 1993 (139); Goddard, 1995 (144)
	6	c.587A>C	p.Tyr178Ser	LS	Korean	Oh, 1999 (159)
	7	c.677A>G	p.Tyr208Cys	LS	Swedish	Enberg, 2000 (160)
	7	c.685C>G	p.Arg211Gly	LS	Mediterranean	Amselem, 1993 (139)
	7	c.686G>A	p.Arg211His	ISS	Iraqi Jewish	Goddard, 1995 (144)
	7	c.718T>C	p.Tyr222His	LS	Not reported	Tauber, 1998 (161)
	7	c.726G>C	p.Glu224Asp	ISS	Not reported	Goddard, 1995 (144)
	7	c.731G>T	p.Ser226Ile	LS	Not reported	Jorge, 2004 (162)
	7	c.784G>A	p.Asp244Asn	LS	Swedish	Enberg, 2000 (160)
	10	c.1319G>T	p.Cys422Phe[b]	LS	Japanese	Kou, 1993 (163); Iida, 1999 (164)
	10	c.1486G>A	p.Ala478Thr	LS, ISS	Not reported	Goddard, 1997 (165); Saenger, 1999 (166)
	10	c.1735C>A	p.Pro561Thr[b]	LS	Japanese	Kou, 1993 (163); Chujo, 1996 (167)

Protein nomenclature is based on mature GHR peptide numbering, which does not include the 18 amino acid residues of the signal peptide. c, Coding DNA sequence where nucleotide 1 is the A of the ATG-translation initiation codon and the transcript includes exon 3; ISS, idiopathic short stature; LS, Laron syndrome; p, protein sequence; X, stop codon.

[a] This mutation was reported as Ser65His.

[b] This amino acid change has also been described in the normal population.

tor site of intron 7 is worthy of mention (54). The effect of the mutation was to impair the anchoring of the variant GHR to the cell membrane. Thus, its continual secretion explained the elevated GHBP, and it is likely that the mutation contributed to the growth disturbance of both child and mother.

C. *STAT5B* mutations

STAT5B mutations present a characteristic phenotype combining GHI and immunodeficiency (OMIM no. 245590). The binding of GH to the GHR activates signaling cascades that include a number of the STAT pathways (STAT1, STAT3, STAT5a, and STAT5b). Molecular defects in GH signal transduction pathways appear to be very rare. However, recent identification of human *STAT5B* mutations causing severe growth failure, as well as marked IGF-I deficiency and GHI, demonstrated that STAT5b signaling is critical for GH-induced IGF-I production and normal linear growth in humans (55). Characteristics of human *STAT5B* mutations are shown in Table 6.

In 2003, the first *STAT5B* mutation was identified in a 16-yr-old female from a consanguineous Argentine family (56). Subsequent reports confirmed that birth weight in affected patients is generally normal but is followed by severe postnatal growth failure with resistance to GH therapy (57–60). The biochemical profile shows normal or

TABLE 5. Splice mutations and insertions/deletions reported in the *GHR* of patients with GHI

Mutation	Exon	cDNA position and nucleotide change	Protein change (mRNA change)	Phenotype	Ethnic origin	First author, year (Ref.)
Splice site	2	c.70+1G>A	p.? (exon 2 skipping)	LS	Turkish	Sobrier, 1997 (142)
	2	c.70+1dupG	p.Ala6GlyfsX7	LS	Turkish	Arman, 2008 (168)
	4	c.266+1G>A	p.Asn28ArgfsX41 (exon 4 skipping)	LS	European, Japanese	Amselem, 1993 (139); Otsuka, 1997 (169)
	6	c.440-1G>C	p.Val129AspfsX18 (exon 6 skipping)	LS	Mediterranean	Amselem, 1993 (139)
	6	c.594A>G	p.Val181_Met188del (aka E180 splice)	LS	Ecuadorian	Amselem, 1993 (139); Berg, 1992 (170)
	6	c.618+792A>G	p.Met188_Met189ins36 (aka pseudoexon activation)	LS	Pakistani	Metherell, 2001 (46)
	7	c.619-1G>T	p.Met189IlefsX8 (exon 7 skipping)	LS	Not reported	Berg, 1993 (140)
	7	c.723C>T	p.Gly223_Glu243del	LS	Spanish, Bahamian	Sobrier, 1997 (142); Baumbach, 1997 (171)
	8	c.785-6T>A	p.Asp244GlyfsX5 (exon 8 skipping)	LS	Bangladeshi	David, 2010 (172)
	8	c.785-3C>A	p.Asp244GlyfsX5 (exon 8 skipping)	ISS	Not reported	Aalbers, 2009 (54)
	8	c.785-1G>T	p.Asp244GlyfsX5 (exon 8 skipping)	LS	Druse, Peruvian	Shevah, 2004 (152); Shevah, 2002 (173)
	8	c.875-1G>C	p.Asp244GlyfsX5 (exon 8 skipping)	LS	Pakistani	Woods, 1996 (174)
	9	c.876-1G>C	p.Ile275LysfsX4 (exon 9 skipping)	ISS	Caucasian	Ayling, 1997 (50)
	9	c.945+1G>A	p.Ile275LysfsX4 (exon 9 skipping)	LS	Japanese	Iida, 1998 (51)
Gross deletions	5	c.267_439del	p.Arg71SerfsX47 (exon 5 skipping)	LS	Sri Lankan	Besson, 2004 (175)
	3, 5, 6	c.71_136del, 267_618del	p.Ala6_Asn28delins Asp, Asn72TrpfsX5 (exons 3, 5, 6 missing)	LS	Jewish-Oriental	Meacham, 1993 (176)
	5	c.267_439del	Arg71SerfsX47 (exon 5 skipping)	LS	Cambodian	Gastier, 2000 (177)
	5, 6	c.267_618del	Asn72TrpfsX5 (exons 5, 6 missing)	LS	Oriental Iraqi/ Iranian Jewish	Godowski, 1989 (6)
	4–10	c.137_1917del	p.? p.28_638del (exons 4–10 missing)	LS	Japanese	Yamamoto, 2008 (178)
Small deletions/ insertions	4	c.161-162delC	p.Lys37SerfsX25	LS	Slovenian	Sobrier, 1997 (142)
	4	c.189-193delTT	p.Ser47MetfsX6	LS	Spanish	Berg, 1993 (140)
	5	c.422insTT	p.Val125LeufsX5	LS	Not reported	Gennero, 2007 (179)
	7	c.742-744del2	p.Tyr230CysfsX12	LS	South African black	Berg, 1993 (140)
	7	c.744delT	p.Tyr230X	LS	Jewish-Iranian	Shevah, 2004 (152)
	9	c.895-899dupC	Val283SerfsX7	LS	Pakistani	Aisenberg, 2010 (53)
	9	c.899-911del13	p.Pro282GlnfsX16	LS	Caucasian	Gastier, 2000 (177)
	10	c.981-982delC	p.Ile310PhefsX21	LS	Not reported	Kaji, 1997 (148)
	10	c.1323-1344del22	p.Ala424SerfsX27	LS	Spanish	Milward, 2004 (28)
	10	c.1733_1734delG[a]	p.Arg560SerfsX23	LS	Not reported	Tiulpakov, 2005 (143)

Protein nomenclature is based on mature GHR peptide numbering, which does not include the 18 amino acid residues of the signal peptide. c, Coding DNA sequence where nucleotide 1 is the A of the ATG-translation initiation codon and the transcript includes exon 3; fs, frameshift; ISS, idiopathic short stature; LS, Laron syndrome; p, protein sequence; p.0? or p.?, the consequences of the mutation at protein level are unknown; X, stop codon.

[a] This mutation was reported as 1776delG in codon 560 according to the old *GHR* numbering.

elevated GH secretion, normal GHBP values, and severe deficiencies of IGF-I, IGFBP-3, and ALS that fail to increase on GH stimulation (57–60). A key feature in all but one reported case (60) was immune dysfunction. In several patients, repeated pulmonary infections occurred from infancy, including episodes of lymphoid interstitial pneumonia (56, 57), a condition associated with autoimmune disease (61). In support of the *in vivo* observations, studies employing both primary fibroblasts from an affected patient (56) and reconstitution systems demonstrated that GH activated STAT5b preferentially over the STAT5a protein, and this specific activation correlated with the induction of IGF-I and IGFBP-3 expression (62). Interestingly, interferon-γ, a cytokine that signals predominantly

TABLE 6. Phenotypes of patients with STAT5b deficiency

Observation	Kofoed, 2003 (56)	Hwa, 2005 (59)	Bernasconi, 2006 (58)	Boyanovsky, 2009 (71)	Vidarsdottir, 2006 (60)	Hwa, 2007 (57)		Martinez, 2007 (70)	Pugliese-Pires, 2010 (69)	
						Sibling 1	Sibling 2		Sibling 1	Sibling 2
Consanguinity	Yes	Yes	NA	Adopted	NA	Yes	Yes	Adopted	NA	NA
Paternal height SDS	−0.3	−0.9	−2.2	NA	−0.8	−1.28	−1.28	NA	−1.5	−1.5
Maternal height SDS	−1.2	−0.6	−3.3	NA	−2.8	−0.6	−0.6	NA	−1.0	−1.0
Sex of proband	F	F	F	F	M	F	F	F	M	M
Age (yr)	16.5	16.4	15.3	12	31	2	4	14.8	6	2
Height SDS	−7.5	−7.8	−9.9	−5.3	−5.9	−5.8	−5.6	−5.95	−5.6	−3
Puberty	Delayed	Delayed	Delayed	Delayed	Delayed	NA	NA	Normal	Delayed	NA
CPD	Yes	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes
Atopy	Eczema	Eczema	Eczema	Eczema	Ichthyosis			Eczema	Atopic	Atopic
Other		Bleeding diathesis		Thyroiditis	Sickle cell anemia	JIA		Thyroiditis		Thrombocytopenic purpura
Hormonal evaluation										
GH basal (ng/ml)	9.4	14.2	6.6	1.8	0.13	17.7	5.7	NA	1.7	1.0
GH stimulated (ng/ml)	53.8	NA	NA	12.5	14.2	NA	NA	27.1	20.6	14
IGF-I (ng/ml)[a]	38	7.0	<10	0.8	14-17	<5	<5	<5	34	<25
IGF-I, GH-stimulated (ng/ml)[a]	55	NA	NA	0.8	78[b]	NA	NA	12	48	<25
IGFBP-3 (mg/liter)[a]	0.87	0.54	NA	0.5	0.18	0.7	0.8	0.84	0.52	0.75
ALS (mg/liter)[a]	2.9	1.2	NA	0.7	0.7	0.4	0.8	NA	NA	NA
Prolactin (μg/liter)	>102 (H)	NA	169 (H)	13–15	110 (H)	NA	NA	83 (H)	61.1 (H)	76.6 (H)
Molecular defect	p.A630P	*c.1191insG*	p.R152X	p.R152X	*c.1102insC*	*c.1680delG*	*c.1680del G*	p.F646S	*c.424_427del*	*c.424_427del*

Homozygous *STAT5B* mutations were identified in 10 case reports. Birth weight and length (data not shown) were normal for gestation. Height SDS (of probands) is at first observation or as reported. Molecular defects encompass missense and nonsense mutations (nomenclature, protein designation) and frameshifts due to insertion or deletion of exonic nucleotides (nomenclature, in *italics*, based on cDNA). c, Coding DNA sequence where nucleotide 1 is the A of the ATG-translation initiation codon; F, female; M, male; H, high, above normal of 20 μg/liter; CPD, chronic pulmonary disease; JIA, juvenile idiopathic arthritis; NA, not available; p, protein sequence; X, stop codon.

[a] All reported values are significantly below the normal range (methodology varied from site to site).

[b] Value determined after 7 d of daily GH injections (at 50 μg/kg body weight).

through the STAT1 pathway, could also up-regulate IGF-I expression, but only if STAT5b was present and fully functional (62). The resulting STAT5b deficiency not only abrogated GH-induced regulation of IGF-I production (63), but IL-2-induced expression of $CD25^{high}$ was also demonstrated to be aberrant (64).

The unusual combination of GHI and features of immune dysfunction in the patient from Argentina led to the identification of a homozygous, missense mutation in exon 15 of *STAT5B* (56). The single *G* to *C* transversion at nucleotide 1888 of the *STAT5B* mRNA resulted in an Ala630 (GCT) to Pro (CCT) substitution. Homology modeling with the solved structure of STAT1 (65) showed that this mutation was within the critical src-homology 2 (SH2) domain, a well-characterized, conserved, regulatory module that functions by interacting with high affinity to phosphotyrosine-containing target peptides. The Ala630Pro (*p.A630P*) substitution was predicted to cause loss of thermodynamic stability as well as aberrant folding and aggregation of the mutant STAT5b protein (66). It is of note that pathogenic mutations in the SH2 domain of a number of cytoplasmic signaling peptides, including STAT1 (67), have been associated with immunodeficiencies (68), but except for STAT5b, none has been associated with severe growth retardation and IGF-I deficiency. Details of the human *STAT5B* mutations reported to date are shown in Fig. 2.

Since the first report, six other human *STAT5B* gene mutations have been documented, with two of the mutations, *c.1680delG* (57) and *c.424_427del* (69), found in siblings. In contrast to children who carry mutations in *IGF1* (see *Section III.E*), brain development and cognitive functions appeared to be normal. The seven *STAT5B* mutations, located in different domains of the STAT5b protein, comprise two missense mutations, *p.A630P* (56) and *p.F646S* (70); a nonsense mutation, *p.R152X* (58, 71); single nucleotide insertions, *c.1191insG* (59) and *c.1103insC* (60); and nucleotide deletions, *c.1680delG* (57) and *c.424_427del* (69). The insertion/deletion mutations result in frameshifts and truncation of the STAT5b protein. All reported mutations were homozygous and autosomal recessive, suggesting that haploinsufficiency of *STAT5B* has minimal impact on IGF-I expression and on growth. The phenotype of STAT5b-deficient patients includes profound short stature and delayed puberty in older subjects. The only patient without obvious immune deficiency, the

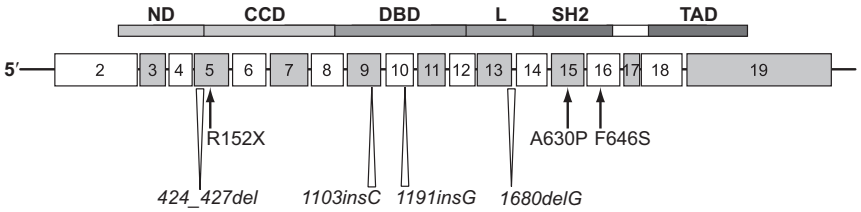

FIG. 2. Homozygous *STAT5B* mutations identified in patients who were GH insensitive, IGF-I deficient, severely growth retarded, and immune compromised. Schematic of the STAT5b protein modular structure encoded by corresponding exons is as indicated. CCD, Coiled-coiled domain; DBD, DNA binding domain; L, linker; ND, N-terminal domain; TAD, transactivation domain.

first reported male proband in the cohort, contracted hemorrhagic varicella at 16 yr of age and had congenital ichthyosis and erythema, but otherwise he appeared relatively healthy (60). One other patient was diagnosed with juvenile idiopathic arthritis at an unusually young age of 2 yr (57). Immunological evaluations reported for one of the patients carrying the *p.R152X* mutation (58) indicated lymphopenia and abnormally low regulatory T cells, similar to the proband carrying the *p.A630P* mutation (56, 64). It is also of note that STAT5b deficiency was associated with abnormally high levels of circulating prolactin in six of the cases (Table 6). It remains unclear whether the hyperprolactinemia is a direct or indirect consequence of *STAT5B* mutations.

D. Mutations of *SHP-2* (encoded by *PTPN11*)

Noonan syndrome (NS) (OMIM no. 163950) and Noonan-like syndromes, which are associated with short stature, are disorders of dysregulation of the rat sarcoma viral oncogene homolog (RAS)-MAPK signaling pathway. In NS, heterozygous mutations in four genes of this pathway (*PTPN11*, *SOS1*, *RAF1*, and *KRAS*) are responsible for approximately 70% of cases (72, 73). The commonest mutation, present in approximately 50% of cases, is in protein tyrosine phosphatase, nonreceptor type 11 (*PTPN11*), which codes for the cytoplasmic tyrosine phosphatase SH2 domain phosphatase-2 (SHP-2), which regulates GH signaling (74) by dephosphorylating STAT5b, causing down-regulation of its activity (75). SHP-2 binds to and dephosphorylates signaling molecules that are positive regulators of the cellular response to GH. Therefore, gain-of-function mutations of *PTPN11* may negatively regulate GH action and thus induce some degree of GHI at a post-GHR level. In support of GHI, serum IGF-I levels were decreased in subjects with NS carrying *PTPN11* mutations (76, 77). There are also data suggesting that *PTPN11* mutation-positive subjects show decreased growth responses to human GH therapy compared with mutation-negative subjects (78).

E. *IGF1* mutations

The first human *IGF1* gene defect was described in 1996 by Woods *et al.* (37) (OMIM no. 608747). Characteristics of patients with *IGF1* defects are shown in Table 7. The first patient, a male, was born by cesarean section because of poor fetal growth. Placental weight was dimin-

TABLE 7. Characteristics of six cases with *IGF1* defects

Observation	Woods, 1996 (37)	Bonapace, 2003 (81)	Walenkamp, 2005 (82)	Netchine, 2009 (83)	van Duyvenvoorde, 2010 (87)	van Duyvenvoorde, 2010 (87)
Sex	M	M	M	M	F	M
Consanguinity	Yes	Yes	Yes	Yes	No	No
Birth weight (SDS/g)	−3.9/1400	−4.0/1480	−2.5/1420	−2.5/2350	−2.9/2300	−1.2/3300
Birth length (SDS/cm)	−5.4/37.8	−6.5/41.0	−3/39.0	−3.7/44.0	−3.8/44.0	−1.0/50.0
Cranial circumference (SDS/cm)	−4.9/27.0	−7.5/26.5	−8.0/44.2	−2.5/32.0	−2.4/47.8	−1.6/49.0
Growth (SDS)	−6.9 at 16 yr	−6.2 at 1.6 yr	−9 at 55 yr	−4.5 at 3 yr	−4.1 at 8.2 yr	−4.6 at 6.2 yr
Microcephaly	Yes	Yes	Yes	Yes	Yes	Mild
Development delay	Yes	Yes	Yes	Mild	Yes	No
Deafness	Yes	Yes	Yes	No	No	No
Adiposity	Yes	No	Yes	No	No	No
Hormonal evaluation						
IGF-I levels	Undetectable	1.0 ng/ml	+7.3 SDS	Variable	−2.3 SDS	−2.6 SDS
IGFBP-3 levels	3.3 mg/liter	3.6 mg/liter	1.98 mg/liter (+0.1 SDS)	4.3 mg/liter	+1.2 SDS	+0.1 SDS
Molecular defect	Hom p.? (Del ex 4–5)	Hom p.?[a]	Hom p.V44M	Hom p.R36Q	Het *c.243-246dupCAGC*	Het *c.243-246dupCAGC*
IGF1R affinity	Zero	Not studied	Extremely low	Partially reduced	Not studied	Not studied

F, Female; M, male; Het, single heterozygous defect; Hom, homozygous defect; p.0? or p.?, the consequences of the mutation at protein level are unknown.

[a] This mutation is localized within the polyadenylation site and alters mRNA splicing. The 3′ end of the resulting aberrant IGF-I transcript contains a partial sequence from the downstream gene *KIAA0537*.

TABLE 8. Phenotypes of patients with homozygous or compound heterozygous *IGFALS* mutations

Observation	Domené, 2004 (26)	Hwa, 2006 (180)	Domené, 2007 (94)			Heath, 2008 (97)		
			Sib 1	Sib 2	Sib 3			
Consanguinity	NA	Yes	No	No	No	No	No	No
Paternal height SDS	NA	−2.0	+1.5	+1.5	+1.5	−0.1	−2.0	−0.4
Maternal height SDS	NA	−2.8	−0.9	−0.9	−0.9	+0.7	−0.6	−0.4
Sex of proband	M	M	M	M	F	M	M	M
Age (yr)	14.6	13.9	15.3	19.6	15.4	4.5	4.6	15.0
Height SDS	−2.1	−2.0	−2.0	−0.5	−1.0	−2.4	−3.9	−2.6
Puberty (yr)	15.7	13.0	16.0	16.9	13.0	12.4	13.5	14.5
Adult height SDS	−1.6	NA	−0.5	−0.5	NA	−2.5	NA	−1.3
Hormonal evaluation								
GH basal (ng/ml)	4.5	1.2	0.2	6.8	NA	3.2	3.9	NA
GH stimulated (ng/ml)	31.0	23.4	10.0	NA	NA	20.7	27.0	6.7
IGF-I (ng/ml)[a]	31	25	8	10	14	15	11	40
IGF-I, GH-stimulated (ng/ml)[a]	39	NA	10	NA	NA	NA	NA	NA
IGFBP-3 (mg/liter)[a]	0.22	0.49	0.38	0.39	0.43	0.70	0.75	0.50
ALS (mg/liter)[a]	<0.5	<0.4	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5
Fasting insulin (μIU/ml) (NR for age)	NA	NA	NA	NA	NA	26.5 (4–11)	40.3 (4–11)	18.7 (4–16)
HOMA (NR <2.6)	NA	NA	NA	NA	NA	4.4	6.2	NA
Mutation	*c.103delG*	p.D440N	p.C540R/p.S195_197Rdup	p.C540R/p.S195_197Rdup	p.C540R/p.S195_197Rdup	p.N276S	p.N276S	p.Q320X

Height SDS (of probands) is at first observation or as reported. Molecular defects encompass missense and nonsense mutations (nomenclature, protein designation) and frameshifts due to insertion or deletion of exonic nucleotides (nomenclature, in *italics*, is based on coding cDNA). F, Female; M, male; SGA, small for gestational age; Sib, sibling; NA, not available; p, protein sequence; X, stop codon; HOMA, homeostasis model of assessment; NR, normal range. Puberty is expressed as onset in males or menarche in females.

[a] All reported values are significantly below the normal range (methodology varied from site to site).

ished (350 g), and he had severe intrauterine growth retardation (IUGR) with a birth weight of 1.4 kg (−3.9 SDS), birth length of 37.8 cm (−5.4 SDS), and microcephaly (head circumference, 27 cm; −4.9 SDS). His growth failure worsened postnatally; at 15.8 yr, his height was 119.1 cm (−6.9 SDS), and his weight was 23.0 kg (−6.5 SDS). He had delayed psychomotor development and sensorineural deafness. During adolescence, he became insulin resistant. No IGF-I was detected in the serum, even after 4 d of stimulation with GH in an IGF-I generation test (IGFGT). Spontaneous 12-h GH secretion showed abnormally elevated baseline and peaks. ALS and IGFBP-3 values were normal. Molecular analysis revealed a homozygous deletion of exons 4 and 5 of the *IGF1* gene. If translated, the resulting protein would be severely truncated, lacking 45 of the 70 IGF-I amino acids (37). At 16.1 yr (bone age, 14.2 yr), rhIGF-I therapy was initiated and resulted in beneficial effects on insulin sensitivity, body composition, bone size, and linear growth (79, 80).

Features of IUGR, microcephaly, retarded intellectual development, and severe postnatal growth failure were present in the other cases with homozygous *IGF1* mutations (81–83). Deafness was present in all the cases except the child with the mildest phenotype (83). The microcephaly that these patients had is a cardinal feature of the phenotype and allows a distinction with patients with Russell-Silver syndrome also born with a severe IUGR, but who have a relative macrocephaly (84). There has been some variation of serum IGF-I levels in the reported *IGF1* mutation cases. The third case to be described (82) shared an identical clinical phenotype with the index case (37) and had a younger brother with similar features who died in childhood (85). This patient had a serum IGF-I level that was significantly increased (+7.3 SDS), which was explained by the fact that the patient's homozygous missense mutation of *IGF1*, a G→A nucleotide substitution at position 274, changing valine at position 44 in the A domain of the mature IGF-I protein to methionine (*p.V44M*), resulted in a recombinant protein (IGF-I V44M) that allowed normal binding to IGFBP-3 but decreased affinity (90-fold) for its receptor, IGF1R (82). This patient therefore had bioinactive IGF-I caused by an *IGF1* mutation. The family members who were heterozygous carriers of this mutation were shorter and had lower head circumferences than the noncarrier family members. Serum IGF-I levels in the fourth patient, who had a relatively mild phenotype, were also variable and not severely decreased (83). In all reported subjects, serum IGFBP-3 and ALS levels have been normal or elevated.

Genetic analyses of *IGF1* in the second case (81) showed a homozygous T→A transversion in exon 6 that would result if translated into an altered E domain of the IGF-I precursor. It has been argued that this variant may be a polymorphism and thus not causative of the patient's phenotype, which was nevertheless strikingly similar to the other cases (86). In the patient with the mild phenotype, sequencing of *IGF1* revealed a homozygous missense mutation resulting in the change of a highly conserved arginine located in the C domain of the protein into a glutamine (*p.R36Q*). Affinity for the IGF1R decreased 2- to 3-fold, resulting in decreased IGF1R autophosphorylation. This partially diminished

van Duyvenvoorde, 2008 (181)											
Sib 1	Sib 2	Sib 3	Fofanova-Gambetti, 2009 (182)				Bang, 2009 (183)		Gallego-Gomez, 2009 (184)	David, 2010 (96)	
Yes	Yes	Yes	NA	No	No	No	No	No	No	No	Yes
−3.2	−3.2	−3.2	NA	+0.3	−1.4	+0.8	NA	NA	NA	0.0	−1.8
−3.4	−3.4	−3.4	NA	−2.1	−0.1	−1.7	NA	NA	NA	−2.0	−1.8
M	M	M	M	F	M	M	M	F	M	M	M
14.6	6.8	4.2	6.7	4.1	15.2	12.7	12.5	11.5	4.1	10.6	13.4
−4.2	−3.0	−3.2	−2.8	−2.1	−3.2	−2.9	−2.4	−2.5	−2.1	−2.8	−3.2
14.0	13.0	13.0	10.5	NA	NA	NA	NA	NA	NA	NA	14.0
−4.2	−3.6	NA	NA	NA	NA	NA	NA	NA	NA	NA	−3.8
NA	NA	NA	NA	NA	NA	NA	NA	NA	69.0	NA	NA
21.0	NA	39.3	25.5	16.2	64.5	63.0	27.0	19.4	<25.0	NA	NA
16	13	44	49	13	37.4	23	−5.0 SD	−6.3 SD	NA	33	40
60	NA	52	NA	NA	38.2	45	NA	NA	<0.5	NA	NA
0.09	0.09	0.10	0.39	0.30	0.50	0.40	−12.7 SD	−13.5 SD	NA	0.20	0.40
<0.07	<0.07	<0.07	<0.1	<0.1	NA	NA	NA	NA	NA	0.3	<0.5
12	17	14	NA	NA	NA	NA	NA	NA	NA	NA	NA
NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
c.1490dupT	*c.1490dupT*	*c.1490dupT*	p.L437_ L439dup	p.C60S/ p.L244F	p.L134Q	p.P73L/ p.L241P	p.L172F/p.S195-197Rdup	p.L172F	p.N276S/*c.184-185insG*	*p.*L134Q/*c.546-548delGGCinsAG*	p.P73L

IGF-I activity had marked consequences for fetal growth and development (83).

A recent publication has reported a definite short stature phenotype associated with a novel heterozygous mutation of the *IGF1* gene (87) (Table 7). Two children, their mother, and their maternal grandfather carried the same mutation—a heterozygous duplication of four nucleotides, resulting in a frameshift and a premature termination codon. The two index cases had severe short stature (height SDS, −4.1 and −4.6), microcephaly, and low IGF-I levels. Adult height of carriers of the maternally transmitted mutation was −2.5 SDS and of the family noncarriers, −1.6 SDS. These data support the hypothesis, previously suggested from larger family studies (82), that heterozygosity for certain *IGF1* mutations may lead to significant short stature. The possibility of the abnormal peptide acting in a dominant negative manner by interfering with IGF1R signal transduction has recently been discounted because the synthetic mutant IGF-I peptide did not bind to the IGFIR or antagonize the growth-promoting effect of IGF-I (88). The spectrum of *IGF1* defects was recently further broadened by the report of a child with short stature (height, −4.0 SDS) and a dominant pattern of inheritance who had IGF-I deficiency and a novel heterozygous donor splice site mutation at the exon 4–intron 4 junction of *IGF1* (89).

F. *IGFALS* mutations

IGFALS mutations are associated with GHI and severe ALS and IGF-I deficiencies (OMIM no. 601489). Characteristics of patients with *IGFALS* mutations are shown in Table 8. The ALS is a soluble protein and a member of the leucine-rich repeat family, and it is expressed by hepatocytes and secreted into the bloodstream (90). GH is the main inducer of ALS synthesis (91), and in the circulation ALS can be found free or bound to IGF-I or -II and IGFBP-3 or -5 to form a ternary complex (92), which prevents IGF, free or bound to IGFBPs, from leaving the circulation, thus prolonging their half-lives and decreasing their availability at a tissue level (93). ALS is encoded by the *IGFALS*, located on chromosome 16p13.3 and spanning 3.3 kb. Inactivation of the *Igfals* gene in mice results in the absence of circulating ALS but only modest growth failure, despite marked reduction of serum IGF-I and IGFBP-3 levels (25).

The first patient with a homozygous mutation of *IGFALS* was reported by Domené *et al.* (26) in 2004 and presented a new combination of genetic, biochemical, and phenotypic data. The most striking feature of this genetic defect causing GHI was a mismatch between extreme deficiencies of circulating IGF-I, IGFBP-3, and ALS and relatively mild growth failure, even leading to a normal adult height in some patients (94). A recent review of published cases confirms these features (95). Sixteen different mutations of the human *IGFALS* gene (Fig. 3) have been identified in 21 cases (95). Eleven were homozygous, six were compound heterozygous, and family studies confirmed autosomal recessive inheritance. *IGFALS* mutations have included missense and nonsense mutations, deletions, duplications, and insertions resulting in

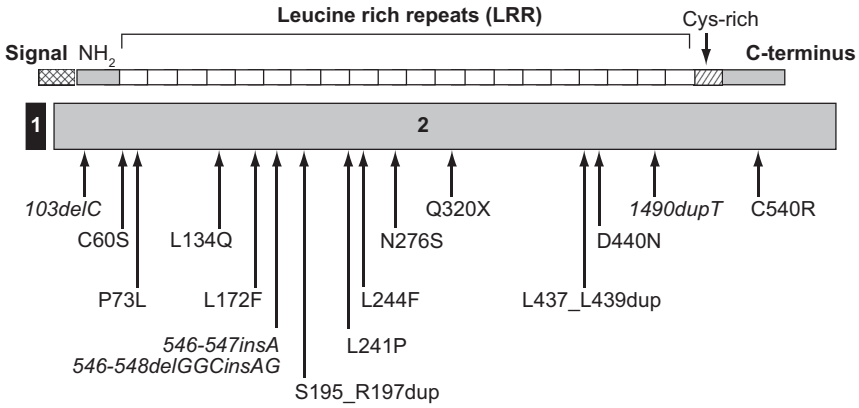

FIG. 3. Schematic representation of the ALS protein indicating the location of identified human *IGFALS* mutations. The *IGFALS* gene is composed of two exons, with five amino residues of the ALS signal peptide encoded by exon 1 and the remainder of the ALS protein encoded by exon 2. NH_2, N-terminal region.

frameshift and premature stop codons and in-frame duplication mutations leading to insertion of extra amino acid residues (95). In all cases, there was extreme deficiency of circulating ALS, with inability to form the ternary complex (95, 96). Whereas circulating levels of IGF-I and IGFBP-3 are severely reduced due to their rapid clearance, local production of IGF-I in peripheral tissues, notably the growth plate, appears to be preserved or even increased due to up-regulation of GH secretion (26). Insulin resistance, with hyperinsulinemia and low IGFBP-1, has also been described in these patients (95, 97).

Recently, attention has focused on the possible effect of heterozygous *IGFALS* mutations on growth. An analysis of 21 patients with homozygous or compound heterozygous *IGFALS* mutations and their family members who were either heterozygous carriers or homozygous wild-type normal has recently been published (98). Mean height SDS was -2.31 ± 0.87 in patients with the homozygous *IGFALS* mutation. Analyses within individual families showed that heterozygosity for *IGFALS* mutations resulted in approximately 1.0 SDS height loss in comparison with wild-type, whereas homozygosity or compound heterozygosity resulted in a further height loss of 1.0–1.5 SDS, suggestive of a gene–dosage effect.

G. *IGFIR* mutations

IGFIR mutations are characterized by IGF-I resistance causing impaired fetal and postnatal growth (OMIM no. 270450). Characteristics of patients with heterozygous *IGFIR* mutations are shown in Table 9. The IGF1R is a transmembrane receptor and belongs to the insulin receptor family, which includes the IGF2R and insulin receptor. The IGFIR is expressed widely and binds IGF-I and -II with high affinity, mediating their biological actions by activating a complex intracellular signaling cascade leading to the transcription of IGF target genes. The *IGF1R* gene is located on chromosome 15q26.3 and spans 315 kb.

TABLE 9. Phenotypes of patients with heterozygous or compound heterozygous *IGFIR* mutations

	Abuzzahab, 2003 (99)				Kawashima, 2005 (100)		Walenkamp, 2006 (39)	
Observation	**Index case**	**Mother**	**Step-brother**		**Index case**	**Mother**	**Daughter**	**Mother**
Consanguinity	No	No	No	No	No	No	No	No
Paternal height SDS	0.1	−2.4	−0.3	−2.8	−2.2	NA	NA	−1
Maternal height SDS	−2.6	−0.8	−2.6	−1.6	−2.9	NA	−4	−1.3
Sex of proband	M	F	M	F	F	F	F	F
Age (yr)	5.3	NA	NA	4.5	6	35	1	31
Birth weight (SD)	2000 g (−3.5)	2400 g (−2.4)	2100 g (−2.7)	1400 g (−3.5)	2686 g (−1.5)	2500 g (−1.6)	2100 g (−3.3)	2600 g (−2.1)
Height SDS	−2.6	−2.6	NA	<−4.0	−2.1	−2.9	<−2.0	−4.0
Adult height SDS	NA	−2.6	NA	−4.8	NA	−2.9	NA	−4.0
Puberty	NA	NA	NA	NA	NA	Normal	NA	Delayed
Microcephaly	Yes	NA	NA	NA	No	NA	Yes	Yes
Intellectual delay	Yes	NA	NA	NA	Yes	NA	NA	No
Other	Facial and body dysmorphisms	NA	NA	NA			Facial dysmorphism, failure to thrive in infancy	Failure to thrive in infancy
Hormonal evaluation								
GH basal (ng/ml)	NA	NA	NA	NA	NA	NA	NA	NA
GH stimulated (ng/ml)	5.7	NA	NA	51	NA	NA	70.4	20.67
IGF-I (NR or SD) (ng/ml)	121 to 222 (+1.1 to +2.3 SD)	NA	NA	1130 (191 to 462)	208 (+1.5 SD)	255 (+0.5 SD)	145 (+2.9 SD)	239 (+1.6 SD)
IGF-I, GH-stimulated (ng/ml)	NA	NA	NA	NA	NA	NA	NA	NA
IGFBP-3 (NR/SD) (mg/liter)	2.1 to 3.7 (−0.7 to +1.8 SD)	NA	NA	5.6 (2.1 to 6.2)	2.22 (−1.0 SD)	2.05 (−0.4 SD)	1.9 (+1.0)	2.63 (+0.1 SD)
ALS (NR) (mg/liter)	NA	NA	NA	28 (5.6 to 16.0)	NA	NA	NA	NA
Molecular defect	Het p.R59X	Het p.R59X	Het p.R59X	p.R108Q/p.K115N	Het p.R709Q	Het p.R709Q	Het p.E1050K	Het p.E1050K

Height SDS (of proband) is at first observation or as recorded. F, Female; M, male; NA, not available; NR, normal range; p.0? or p.?, the consequences of the mutation at protein level are unknown.

Mutations in *IGF1R* were first reported in 2003 by Abuzzahab *et al.* (99) after analyses of DNA from cohorts of children with short stature and unexplained IUGR. The first child was a compound heterozygote for point mutations in exon 2 of the *IGFIR* gene that altered the amino acid sequence to *p*.R*108*Q in one allele and *p*.K*115*N in the other. She had a birth weight of −3.5 SDS with childhood short stature and an adult height of −4.8 SDS. The second patient, a boy, had a heterozygous nonsense mutation (*p.R59X*) that reduced the number of IGF1R on fibroblasts. He also had low birth weight (−3.5 SDS) and birth length (−5.8 SDS) with microcephaly and postnatal growth failure (height, −3.8 SDS) at age 14 months and some additional dysmorphic features. Serum IGF-I levels were normal or elevated in both patients, and GH secretion was within normal limits.

There have been several reports of familial short stature due to *IGF1R* mutations. A heterozygous mutation in the cleavage site of the proreceptor of *IGF1R* was reported in a 6-yr-old Japanese girl and her mother (100), and the research team of Wit and colleagues (39) in Leiden described a mother and daughter with a heterozygous missense mutation in the intracellular part of the *IGF1R*. A 9-yr-old male patient (height, −3.6 SDS), his sister (height, −1.94 SDS; and IUGR), and his mother (height, −4.6 SDS) carried the same *IGF1R* mutation, which was a novel heterozygous 19-nucleotide duplication in exon 18 (101). Functional studies in primary dermal fibroblasts derived from the patient and family members indicated that *IGF1R* mRNA expressed from the mutant allele was degraded through the nonsense-mediated mRNA decay pathway, resulting in a reduced amount of wild-type IGF1R protein and, subsequently, diminished activation of the IGF1R pathway. Another female child was reported with a heterozygous missense mutation in the highly conserved N-terminal fibronectin type-III domain of *IGF1R*, with functional studies demonstrating impaired postreceptor IGF-I signaling (102).

Recently, a new mechanism of IGF-I resistance has been proposed from the study of a patient with a *p.V599E IGF1R* mutation, which interfered with receptor trafficking, thereby abolishing proreceptor processing and plasma membrane localization and leading to insufficient IGF-I signaling (103). In another case with a *p.G1125A* mutation, cotransfection of wild-type and mutant *IGF1R* resulted in reduced autophosphorylation of 36 ± 10% of wild-type levels, leading to a kinase-deficient IGF1R (104). It was hypothesized that this partial dominant negative effect would be likely to cause the phenotype of IUGR and postnatal growth retardation. A recent review of the literature demonstrated that small birth size, childhood short stature, small head size, relatively high IGF-I levels, developmental delay, and micrognathia are the main predictors for the diagnosis of an *IGF1R* deletion (105).

H. *GH1* mutations causing biologically inactive GH

Certain *GH1* mutations cause biologically inactive GH, resulting in a form of GHI (OMIM no. 262650). A syndrome of biologically inactive GH causing GHI and

Inagaki, 2007 (102)	Fang, 2009 (101)	Ester, 2009 (105)		Wallborn, 2010 (103)	Kruis, 2010 (104)
No	No	Yes	No	No	No
−2.2	−1.7	0.4	+0.8	178 cm	NA
−5.7	−4.6	1.3	−0.5	−3.3	NA
F	M	F	M	F	F
13.6	9.5	2.3	3	9	7.5
2100 g (−3.1)	2366 g (NA)	2890 g (−1.3)	2600 g (−1.9)	2250 g (−2.3)	2450 g (−1.8)
−5.0	−3.6	−3.5	−3.8	−2.1	−3.4
NA	NA	NA	NA	NA	NA
Normal	NA	NA	NA	NA	NA
No	Yes			Yes	Yes
No	No	Yes		Yes	No
	Same mutation also reported in mother (height −4.6 SD) and sister (height −1.95 SD)	Facial dysmorphism, limb deformity, hearing loss	Facial dysmorphism, limb deformity		
0.4	0.42	NA	NA	NA	0.9
10.6	9.1	6.4	23.2	NA	>40.0
404 (165 to 300)	164 (123 to 275)	34 (−2.78 SD)	208 (+1.25 SD)	285 (+1.83 SD)	200 (+0.76 SD)
354.2	402	197 (+1.91 SD)	356 (+2.28 SD)	357	NA
73.1 (3.5 to 35.0)	2.34 (2.0 to 4.8)	1.35 (−1.33 SD)	2.99 (+1.24 SD)	4.3	NA
NA	NA	NA	NA	NA	NA
Het p.R481Q	Het *c.3348_3366dup*	Het p0.0 (deletion of all exons)	Het p.? (deletion including exons 3–21)	Het p.V599E	Het p.G1125A

short stature was first described by Kowarski *et al.* (106) in 1978. The molecular basis of this apparently rare cause of GHI was clarified by Takahashi *et al.* (107, 108), who reported two cases with heterozygous mutations in the *GH1* gene. The first case was a boy with severe short stature (height, −6.1 SDS) who had increased immunoassayable GH and IGF-I deficiency, which responded as did his growth to exogenous human GH therapy. The mutation was single-base missense substitution (*p.R77G*) in exon 4 of the *GH1* gene. However, his normal-stature father had the same mutation. Functional studies demonstrated that the mutant GH molecule had higher binding affinity for the GHR and inhibited its activation by wild-type GH in a dominant negative fashion, thus impairing GH bioactivity. The second case was a girl with similar endocrine features and short stature associated with a heterozygous single-base change (A→G) causing a p.D112G substitution in *GH1*. More recently, a child with short stature (height at age 9 yr, −3.6 SDS) from a consanguineous Serbian family was reported to have a homozygous missense mutation, Cys53Ser, of *GH1* leading to the absence of the disulfide bridge Cys-53 to Cys-165 in the GH molecule (109). Functional studies demonstrated that GH binding and JAK2/STAT5 signaling pathway activation were significantly reduced in the mutant GH-C53S compared with wild-type-GH. Both growth and serum IGF-I responded well to GH therapy.

I. *GHI* deletions (type IA GH deficiency) with anti-GH antibodies

A rare form of GHI occurs due to acquired GH-inhibiting antibodies in a category of children with familial isolated GH deficiency (IGHD) (OMIM no. 262400) (110). Autosomal recessive IGHD, caused by gross deletions of the *GH1* gene, result in severe IGHD (type IA) with undetectable GH secretion (111). Such patients have severe postnatal growth failure with height usually less than −4.5 SDS. Most of the *GH1* deletions are 6.7, 7.0, or 7.6 kb in length, although several of 45 kb have been reported (112). Microdeletions and frameshift mutations have also been reported (45, 110, 113). Patients with IGHD and homozygous deletions of the *GH1* gene frequently develop anti-GH antibodies during treatment with GH due to immunological intolerance. However, variability of both antibody formation and response to GH therapy may occur, even within families (112). Rare homozygous microdeletions and single base-pair substitutions in the *GH1* coding region have also resulted in anti-GH antibody formation during GH therapy. However, patients who are heterozygous for a *GH1* deletion but whose other *GH1* allele is not deleted and produces a nontruncated product usually do not develop anti-GH antibodies (110). The formation of anti-GH antibodies neutralizes the growth response to GH therapy, resulting in a state of GHI associated with severe short stature. Such patients may respond to therapy with rhIGF-I, which becomes the only effective management for their growth failure (114).

IV. The Continuum of Phenotypic Features

The concept of genotype:phenotype relationships in endocrinology evolved from evidence that a range of genetic defects can influence phenotypic expression based on the degree of disturbance of key endocrine mechanisms. For example, it is well established in congenital adrenal hyperplasia that certain *CYP21* mutations cause different degrees of steroidogenic disruption resulting in a range of clinical phenotypes (115).

This hypothesis can be applied to defects of the GH–IGF axis causing GHI. Since populations of children with GHI were first reported, a range of phenotypes has been described. This was very noticeable in the series of 82 patients, mainly of European origin, who were identified in the early 1990s for rhIGF-I therapy (116). There was a gradation of severity of short stature, with height SDS ranging from −2.2 to −10.4 and a strong positive correlation ($r^2 = 0.45$; $P \leq 0.001$) between height SDS and IGFBP-3 SDS. A further variable in the same population related to phenotype was the serum GHBP level, which when very low or absent was associated with more severe short stature (height SDS, −6.45), whereas normal GHBP values were associated with milder short stature (height SDS, −4.89) (116). A study of craniofacial phenotype in the same group of subjects identified that those with normal facial appearance, as opposed to the dysmorphic features of Laron syndrome, had milder short stature and could present as idiopathic short stature (117). However, in this series of GH-resistant patients, there was no clear relationship between *GHR* mutation and phenotype (116).

In populations of patients from Ecuador with the homozygous E180 splice *GHR* mutation, heterogeneity of statural phenotype was also seen with height SDS values ranging from −5.3 to −11.5, and height SDS correlated positively ($P < 0.01$) with both IGF-I SDS and IGFBP-3 SDS values (8, 118).

A. *GHR* mutations

Thirty-eight patients fulfilling the criteria of GHI (119) were studied in the Centre for Endocrinology at Barts and the London School of Medicine and Dentistry (London, UK) and were identified to have homozygous, compound heterozygous, or heterozygous dominant negative *GHR*

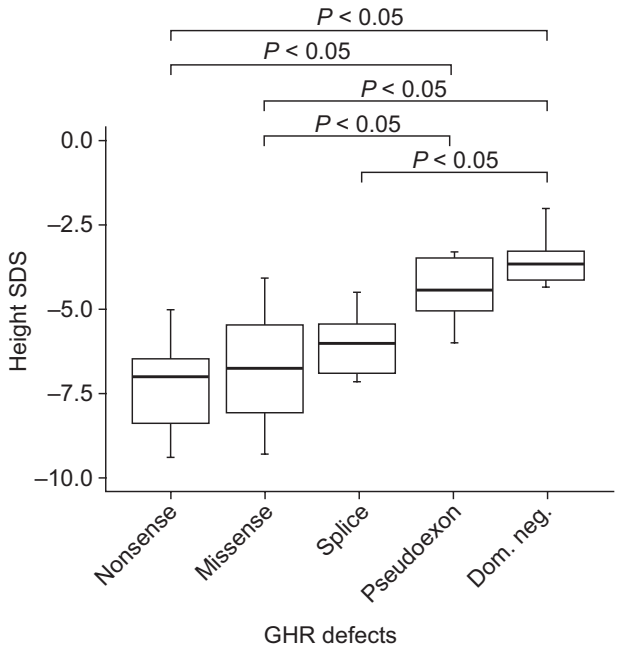

FIG. 4. Height SDS values in 70 children with GHI and *GHR* mutations divided according to the type of mutation. Each *boxplot* depicts the median and the 25th and 75th percentiles. *Whiskers* depict minimum and maximum observed values. Statistical analyses were performed using R version 2.6.2 (R Development Core Team, 2008, R Foundation for Statistical Computing, Vienna, Austria). Numerical variables were expressed as median (range). Comparison between continuous variables was performed using the Student's *t* test. A two-sided *P* value <0.05 was considered indicative of statistical significance. Bonferroni adjustment was performed to reduce the likelihood of type I error. Dom. neg, Dominant negative.

mutations. To perform an assessment of height SDS and type of *GHR* mutation, these 38 subjects were analyzed together with 32 subjects, also fulfilling the same GHI criteria, who were added from the literature (9). Relationships between *GHR* mutation type and height SDS are shown in Fig. 4. It is of interest that, for the first time, dominant negative *GHR* mutations (50, 51) and *GHR* intronic pseudoexon mutations (49) were shown to be associated with significantly less severe growth phenotypes ($P < 0.05$) than *GHR* missense and nonsense mutations. The probable explanation is that in the case of the dominant negative mutations there will still be a proportion of normally functioning receptor dimerized with a second normally functioning receptor, resulting in effective GH signal transduction. In the case of the pseudoexon mutation, there is a small residue of normally spliced receptor that will result in a degree of normal GH signaling.

B. *STAT5B* and *IGFALS* mutations

Patients with homozygous *STAT5B* mutations have a range of phenotypic characteristics, which are shown in Tables 2 and 6. With respect to their growth, height SDS values ranged from −5.6 to −9.9. It must be remembered that the majority of these patients had serious immunological abnormalities, frequently requiring immunosuppressive or steroid therapy, which almost certainly contributed to their growth failure. In a recent review of 17 cases with homozygous *IGFALS* mutations, height SDS values in prepubertal subjects (n = 15) ranged from −1.1 to −3.9 (mean, −2.6), in adolescent subjects (n = 10) from −1.0 to −4.4 (mean, −2.8), and in adult subjects (n = 8) from −0.5 to −4.4 (mean, −2.2) (95) (Table 8). These data emphasize the mildness of the growth defect in *IGFALS* mutation subjects, albeit with variability, and a trend showing improvement in height SDS as age advances.

C. *IGF1* and *IGF1R* mutations

The key feature of *IGF1* and *IGF1R* mutations, not present with other genetic causes of GHI, is their association with impaired fetal growth. Consequently, the combination of fetal and postnatal growth failure is suggestive of these mutations. Key additional phenotypic features of *IGF1* defects are microcephaly, deafness, and some degree of intellectual retardation (38) (Table 7). However, as more cases are diagnosed, the phenotype is likely to evolve. A recent review of the literature of *IGF1R* mutations demonstrated that small head size, relatively high IGF-I levels, developmental delay, and micrognathia were the main predictors for this diagnosis (Table 9) (105).

V. The Continuum of Biochemical Changes

Disturbance of the GH–IGF-I axis is present by definition in all patients with GHI, and their biochemical abnormalities reflect this (9). The cardinal biochemical features of GHI states are deficiency of IGF-I and normal or increased GH concentrations. An exception to IGF-I deficiency is seen with *IGF1R* mutations where IGF-I is usually normal or elevated. In *GHR* mutations, IGF-I, IGFBP-3, and ALS levels are usually severely decreased, although the degree of the deficiencies is variable, supporting the evidence for a continuum. Most homozygous *GHR* mutations cause extreme deficiency of all GH-dependent peptides, associated with increase in basal and stimulated GH secretion (8, 116, 120, 121). However, a range of IGF-I and IGFBP-3 deficiencies was present, particularly in some of the less homogeneous populations such as the European GHI series where subtle differences, particularly in IGFBP-3, being less sensitive to GH, was noticeable (116). A range of IGF-I deficiency was also present in the severely affected patients from Ecuador (118).

Some *GHR* mutations result in detectable, albeit low, IGF-I levels. Patients with dominant negative, splice site, and the pseudoexon 6Ψ mutations have milder IGF-I deficiency compared with GHI subjects with nonsense and

missense mutations. The milder phenotype could be explained by the three possible combinations of GHR dimerization, which can occur in these patients. The presence of the wild-type GHR transcript has, in fact, been documented in subjects with GHI caused by a homozygous pseudoexon 6Ψ mutation (46). For reasons as yet unknown, a splice mutation may not always be 100% efficient in causing aberrant splicing, and the coexistence of normal and mutant transcripts may occur. Therefore, the wild-type/wild-type GHR dimer can be present alongside the nonfunctional wild-type/mutant-type heterodimer and the nonfunctional mutant-type/mutant-type homodimer. The presence of different ratios of mutant to wild-type receptor can also explain the occurrence of different phenotypes within patients with the same splice mutation, as is known to occur when the genetic defect causes aberrant mRNA splicing (122, 123). Another possible reason for the IGF-I variability observed in patients with severe GHI due to different *GHR* mutations can be the influence of puberty and sex steroids. The increased production of estrogen and testosterone during puberty and the resulting increase in GH levels may induce a rise, albeit modest, of IGF-I levels, particularly in patients with the milder phenotype caused by dominant negative, pseudoexon 6Ψ, and splice *GHR* mutations.

A. *GHR* mutations

In the recent study of GHI cases with identified *GHR* mutations performed at St. Bartholomew's Hospital, serum IGF-I levels were available in 41 subjects. A range of IGF-I deficiency was demonstrated in association with different *GHR* mutations. Relationships between type of mutation and IGF-I SDS values are shown in Fig. 5. IGF-I SDS values were significantly lower ($P < 0.05$) in subjects with missense and nonsense *GHR* mutations than in those with the pseudoexon mutations, as previously described (49). Patients with *GHR* splice site mutations and dominant negative defects had significantly higher ($P < 0.05$) IGF-I levels than subjects with missense mutations. These findings show that a continuum of IGF-I levels, the key biochemical abnormality of GHI, exists across the spectrum of *GHR* mutations, linked to the type of mutation and thus reflecting the degree of receptor dysfunction. A similar analysis has not been performed for IGFBP-3 and ALS, but GHBP concentrations were available in 58 of the GHI subjects. GHBP levels, which may be normal depending on the site of the mutation in *GHR* (8, 116), were categorized as undetectable, low, normal, or high, and they varied considerably according to the mutation responsible for the GHI (Fig. 6). They demonstrate another example of a biochemical continuum across the GHI spectrum.

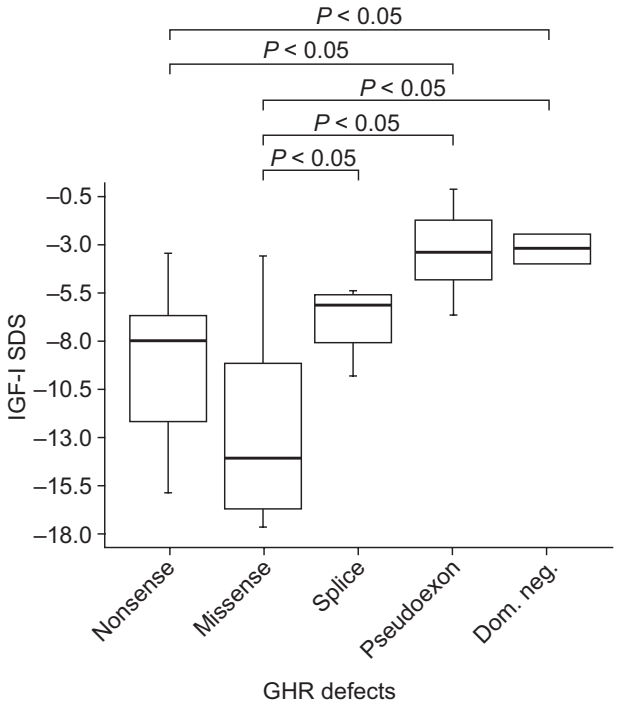

FIG. 5. IGF-I SDS values in 41 children with GHI and *GHR* mutations divided according to the type of mutation. Each *boxplot* depicts the median and the 25th and 75th percentiles. *Whiskers* depict minimum and maximum observed values. Statistical analyses were performed as in Fig. 4. Serum IGF-I levels were measured from venous blood samples using ELISA kit (Diagnostic System Laboratories, Inc., Webster, TX) by Dr. F. Miraki-Moud in the laboratories of Dr. C. Camacho-Hübner at Barts and the London School of Medicine and Dentistry (London, UK). For IGF-I, the assay sensitivity was 0.03 ng/ml. The intra- and interassay coefficients of variation were 8.6 and 6.8% for mean serum concentrations of 104 and 90 ng/ml, respectively. Normal values for IGF-I were obtained from Diagnostic System Laboratories and were used to calculate SDS. IGF-I levels are not normally distributed, so values were converted to their normal logarithm. For the purpose of the analysis, undetectable values were arbitrarily substituted for the values immediately below the assay sensitivity (*e.g.* undetectable IGF-I levels were considered = 0.01 ng/ml). Dom. neg, Dominant negative.

B. *STAT5B* and *IGFALS* mutations

STAT5B mutations are associated with severe GHI, which is reflected in marked deficiencies of all GH-dependent peptides and increase in GH secretion (55). This is summarized in Table 6. In subjects with homozygous *IGFALS* mutations, serum ALS levels are almost universally undetectable, and failure to form the circulating ternary complex results in rapid clearance and extreme deficiency of IGF-I and ALS, as described in the first reported patient (26) and in subsequent case reports (95) (Table 8). In the recent review by Domené *et al.* (95), serum IGF-I SDS values ranged from −3.2 to −11.2 and IGFBP-3 SDS values from −3.6 to −18.5 (95). GH secretion was increased in the majority of subjects, which may contribute to some degree of insulin resistance.

C. *IGF1* and *IGF1R* mutations

In the four cases with homozygous *IGF1* defects and the patients with a heterozygous mutation causing a definite

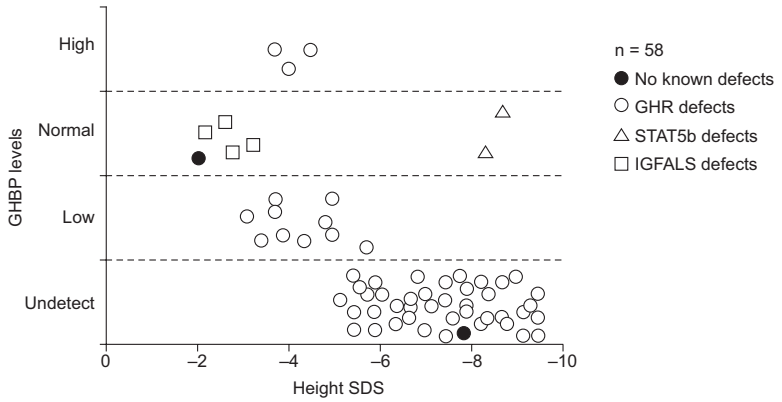

FIG. 6. Serum GHBP concentrations in 58 children with GHI. GHBP values were categorized as: above the normal range, in the normal range, below the normal range, or undetectable, and were plotted according to the height SDS values of the subjects. Subjects with *GHR*, *STAT5B*, and *IGFALS* mutations or no known defects are shown by different symbols. Undetect, Undetectable.

growth phenotype, serum IGF-I levels varied according to the nature of the mutation. IGFBP-3 and ALS levels were notably normal, and GH secretion was usually normal or increased (Table 7) (38). The biochemical characteristics of children with heterozygous *IGF1R* mutations were recently reviewed (105). Essentially, serum IGF-I may be increased or normal, and IGFBP-3 is usually within the normal range.

VI. An Updated Approach to the Investigation of GH Insensitivity

The evaluation of a child with short stature and possible GHI should comply with the classical paradigm of clinical assessment followed by general (*i.e.* nonendocrine) investigations, hormonal assessment, and possible genetic analyses. An algorithm showing key steps in the investigation of genetic GH–IGF-I axis defects is indicated in Fig. 7. As advances in molecular endocrinology related to growth disorders progress, the crucial importance of detailed phenotypic evaluation and documentation becomes increasingly appreciated. The urge to obtain a possible molecular diagnosis at the onset of the investigations should be resisted until detailed clinical and endocrine evaluation has been performed. Clinical assessment should include inquiries about family history of growth disturbance, consanguinity, birth weight and length, and recurrent infections (124). Examination should specifically assess the presence of possible facial dysmorphic features and microcephaly in addition to anthropometric evaluation (125).

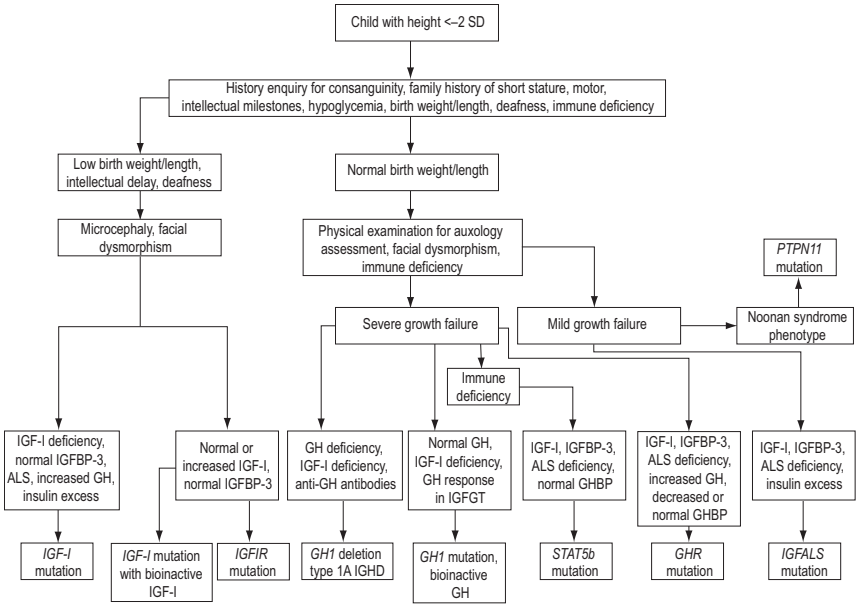

FIG. 7. Algorithm showing key steps in the investigation of genetic GH–IGF-I axis defects.

A. Investigations of the GH–IGF-I axis

Investigations of the GH–IGF-I axis consist of determination of GH secretion and exploration of the IGF system. A GH provocation test is recommended unless the child has normal auxology or a basal IGF-I level above the mean for age (125). In a child with clinical criteria of GH deficiency, a peak GH level below 10 ng/ml has traditionally been used to support this diagnosis (126). Basal IGF-I levels should also be determined, although these may be influenced by factors such as age, nutrition, chronic illness, and puberty. In the initial assessment, IGFBP-3 adds little, except in children under 3 yr of age, where low IGFBP-3 is helpful in the diagnosis of GH deficiency (127). Reliable assay performance and appropriate normative data (128, 129) are essential for the use of IGF-I and IGFBP-3 in clinical practice, and adjustment for sex, age, puberty, and nutritional status is recommended.

A diagnosis of GHI follows from the demonstration of abnormal auxology, normal GH secretion, and IGF-I deficiency. However, the pathogenesis will not have been elucidated from these investigations. The nature of the defect can often be defined by additional measurement of IGFBP-3, ALS, and GHBP (124). In *GHR* defects, IGF-I, IGFBP-3, and ALS are decreased (121), although the degree of abnormality can vary with the type of mutation, which also influ-

ences GHBP levels. GH secretion is elevated in most patients with severe IGF-I deficiency.

B. The IGF-I generation test

The principle behind the design of the IGFGT was that repeated injections of human GH induce measurable increases of IGF-I, IGFBP-3, and ALS secretion. However, in GH-deficient patients, the degree of IGF-I response did not convincingly predict the growth response to GH therapy (130). Normative data were not established, and the test is not used for this purpose. Interest in the IGFGT was renewed when molecular evidence of GHI was demonstrated and subjects were selected for rhIGF-I therapy. Criteria for diagnosis of GHI were defined as: failure to increase IGF-I and IGFBP-3 by more than 15 and 400 ng/ml, respectively (116, 119). However, because the spectrum of GHI disorders expanded, these criteria now appear to be too strict for more mildly affected subjects and even in patients with severe GHI, post-GH increases of IGF-I ranged from below 20 to 58 ng/ml, and of IGFBP-3 from 95 to 1762 ng/ml (131). Attempts to refine the IGFGT for the diagnosis of milder GHI have demonstrated that patients with idiopathic short stature produced a subnormal response (132), and subjects with IGF-I deficiency and normal GH secretion also had subnormal ability to generate IGF-I (133). However, additional sensitivity for the diagnosis of GH resistance was not seen with a low-dose GH protocol (134). A lack of reproducibility of IGF-I and IGFBP-3 responses in the IGFGT has also been reported (135). For this reason, genetic analysis and assessment of the growth response to GH therapy, especially in patients without the classical GHI phenotype, should be performed to confirm GH resistance. The principal value of the IGFGT is the confirmation of extreme or severe GHI (8, 116).

C. Genetic investigations

The discussions above have indicated that genetic mutations in the GH–IGF-I axis make a major contribution to the pathogenesis of GHI. After clinical and biochemical assessment and where a genetic cause of short stature is expected from the family history, DNA analysis for key candidate genes can confirm a genetic diagnosis. A hierarchy and priority of molecular tests can be defined after careful clinical and biochemical assessment. Testing for molecular defects in the GH–IGF-I axis is not commercially available at the present time. However, there is a number of academic laboratories that perform DNA sequencing studies of the relevant candidate genes. *In vitro* functional studies may also be necessary to quantitate the degree of protein dysfunction, particularly in cases with a milder phenotype. There are many components of the GH and IGF signaling cascades that remain poorly understood and are legitimate candidates for harboring significant mutations and/or deletions. Thus, the absence of identifiable mutations in the candidate genes described above cannot exclude the possibility of a molecular abnormality of the GH–IGF axis.

Ideally, family members should also be tested. It is now clear that members of the same family with the same mutations may have differing phenotypes (28). Additionally, for many of the autosomal recessive disorders described above, the issue of heterozygous expression remains of great interest and is worthy of further study (87, 98).

VII. Conclusions and Future Perspectives

From the fundamental importance of the GH–IGF axis in human linear growth, it follows that defects at many points in this axis will result in growth impairment leading to childhood and adult short stature. The key defects leading to GHI have been described, and the range of genetic, clinical, and biochemical abnormalities, both within each genetic disorder and within the spectrum of GHI disorders as a whole, has been emphasized. GHI can no longer be considered to be a single clinical entity, as it was envisaged nearly 50 yr ago.

As new genetic defects leading to an expansion of the field of GHI are described, each new mutation will itself contribute to the genetic and phenotypic continuum. Although the precise etiology in many children with short stature remains uncertain, the investigation of patients with abnormal growth should be encouraged to improve diagnosis and contribute to science. We suggest that the concept of a continuum of defects causing GHI, many with overlapping phenotypes, may help the clinician to consider appropriate investigation and management options.

Acknowledgments

We thank Yves Le Bouc, Salah Azzi, and Muriel Houang in the Institut National de la Santé et de la Recherche Médicale, Unité 938 (Paris, France) for their help in the identification and extensive characterization of the partial *IGF1* defect.

Address all correspondence and requests for reprints to: Professor Martin Savage, Department of Endocrinology, Barts and the London School of Medicine and Dentistry, John Vane Science Centre, Charterhouse Square, London EC1M 6BQ, United Kingdom. E-mail: m.o.savage@qmul.ac.uk.

This work was supported by the Institut National de la Santé et de la Recherche Médicale, Unité 938, Team 4 (to I.N.); Agence Nationale de la Recherche (to I.N.); Université Pierre et Marie Curie, Assistance Publique Hôpitaux de Paris (to I.N.); Direction de l'Hospitalisation et de l'Organistion des Soins/Mission de L'observation, de la Perspective et de la Recherche 243/25.0505 (to I.N.); the Molecular Endocrinology Lab-

oratory in the William Harvey Research Institute (London, UK), and Ipsen (to A.J.L.C.); the IGFD Research Center (Portland, OR) (to R.G.R.); and Ipsen and the March of Dimes (to R.G.R.).

Disclosure Summary: A.D., V.H., L.A.M., I.N., and A.J.L.C. have nothing to disclose. C.C.-H. is currently employed by Pfizer Global Pharmaceuticals (New York, NY). R.G.R. has received consulting fees from Ipsen and Lilly and honoraria from the Advisory Council Inc. M.O.S. received consultancy fees from Ipsen and honoraria from the Advisory Council Inc. and Pfizer Inc.

References

1. **Laron Z, Pertzelan A, Mannheimer S** 1966 Genetic pituitary dwarfism with high serum concentration of growth hormone—a new inborn error of metabolism? Isr J Med Sci 2:152–155
2. **Laron Z** 2004 Laron syndrome (primary growth hormone resistance or insensitivity): the personal experience 1958–2003. J Clin Endocrinol Metab 89:1031–1044
3. **Eshet R, Laron Z, Pertzelan A, Arnon R, Dintzman M** 1984 Defect of human growth hormone receptors in the liver of two patients with Laron-type dwarfism. Isr J Med Sci 20: 8–11
4. **Laron Z, Klinger B, Erster B, Anin S** 1988 Effect of acute administration of insulin-like growth factor I in patients with Laron-type dwarfism. Lancet 2:1170–1172
5. **Walker JL, Ginalska-Malinowska M, Romer TE, Pucilowska JB, Underwood LE** 1991 Effects of the infusion of insulin-like growth factor I in a child with growth hormone insensitivity syndrome (Laron dwarfism). N Engl J Med 324:1483–1488
6. **Godowski PJ, Leung DW, Meacham LR, Galgani JP, Hellmiss R, Keret R, Rotwein PS, Parks JS, Laron Z, Wood WI** 1989 Characterization of the human growth hormone receptor gene and demonstration of a partial gene deletion in two patients with Laron-type dwarfism. Proc Natl Acad Sci USA 86:8083–8087
7. **Amselem S, Duquesnoy P, Attree O, Novelli G, Bousnina S, Postel-Vinay MC, Goossens M** 1989 Laron dwarfism and mutations of the growth hormone-receptor gene. N Engl J Med 321:989–995
8. **Rosenfeld RG, Rosenbloom AL, Guevara-Aguirre J** 1994 Growth hormone (GH) insensitivity due to primary GH receptor deficiency. Endocr Rev 15:369–390
9. **Savage MO, Attie KM, David A, Metherell LA, Clark AJ, Camacho-Hübner C** 2006 Endocrine assessment, molecular characterization and treatment of growth hormone insensitivity disorders. Nat Clin Pract Endocrinol Metab 2:395–407
10. **Salmon Jr WD, Daughaday WH** 1957 A hormonally controlled serum factor which stimulates sulphate incorporation by cartilage *in vitro*. J Lab Clin Med 49:825–836
11. **Green H, Morikawa M, Nixon T** 1985 A dual effector theory of growth-hormone action. Differentiation 29:195–198
12. **Roberts Jr CT, Lasky SR, Lowe Jr WL, Seaman WT, LeRoith D** 1987 Molecular cloning of rat insulin-like growth factor I complementary deoxyribonucleic acids: differential messenger ribonucleic acid processing and regulation by growth hormone in extrahepatic tissues. Mol Endocrinol 1:243–248
13. **Han VK, Lund PK, Lee DC, D'Ercole AJ** 1988 Expression of somatomedin/insulin-like growth factor messenger ribonucleic acids in the human fetus: identification, characterization, and tissue distribution. J Clin Endocrinol Metab 66:422–429
14. **Lowe Jr WL, Roberts Jr CT, Lasky SR, LeRoith D** 1987 Differential expression of alternative 5′ untranslated regions in mRNAs encoding rat insulin-like growth factor I. Proc Natl Acad Sci USA 84:8946–8950
15. **Lowe Jr WL, Lasky SR, LeRoith D, Roberts Jr CT** 1988 Distribution and regulation of rat insulin-like growth factor I messenger ribonucleic acids encoding alternative carboxyterminal E-peptides: evidence for differential processing and regulation in liver. Mol Endocrinol 2:528–535
16. **Isaksson OG, Jansson JO, Gause IA** 1982 Growth hormone stimulates longitudinal bone growth directly. Science 216:1237–1239
17. **Nilsson A, Isgaard J, Lindahl A, Dahlström A, Skottner A, Isaksson OG** 1986 Regulation by growth hormone of number of chondrocytes containing IGF-I in rat growth plate. Science 233:571–574
18. **Isaksson OG, Lindahl A, Nilsson A, Isgaard J** 1987 Mechanism of the stimulatory effect of growth hormone on longitudinal bone growth. Endocr Rev 8:426–438
19. **LeRoith D, Bondy C, Yakar S, Liu JL, Butler A** 2001 The somatomedin hypothesis: 2001. Endocr Rev 22:53–74
20. **Sjögren K, Liu JL, Blad K, Skrtic S, Vidal O, Wallenius V, LeRoith D, Törnell J, Isaksson OG, Jansson JO, Ohlsson C** 1999 Liver-derived insulin-like growth factor I (IGF-I) is the principal source of IGF-I in blood but is not required for postnatal body growth in mice. Proc Natl Acad Sci USA 96:7088–7092
21. **Yakar S, Liu JL, Stannard B, Butler A, Accili D, Sauer B, LeRoith D** 1999 Normal growth and development in the absence of hepatic insulin-like growth factor I. Proc Natl Acad Sci USA 96:7324–7329
22. **Wu Y, Sun H, Yakar S, LeRoith D** 2009 Elevated levels of insulin-like growth factor (IGF)-I in serum rescue the severe growth retardation of IGF-I null mice. Endocrinology 150: 4395–4403
23. **Kaplan SA, Cohen P** 2007 The somatomedin hypothesis 2007: 50 years later. J Clin Endocrinol Metab 92:4529–4535
24. **Ohlsson C, Mohan S, Sjögren K, Tivesten A, Isgaard J, Isaksson O, Jansson JO, Svensson J** 2009 The role of liver-derived insulin-like growth factor-I. Endocr Rev 30:494–535
25. **Ueki I, Ooi GT, Tremblay ML, Hurst KR, Bach LA, Boisclair YR** 2000 Inactivation of the acid labile subunit gene in mice results in mild retardation of postnatal growth despite profound disruptions in the circulating insulin-like growth factor system. Proc Natl Acad Sci USA 97:6868–6873
26. **Domené HM, Bengolea SV, Martínez AS, Ropelato MG, Pennisi P, Scaglia P, Heinrich JJ, Jasper HG** 2004 Deficiency of the circulating insulin-like growth factor system associated with inactivation of the acid-labile subunit gene. N Engl J Med 350:570–577
27. **Derr MA, Fang P, Sinha SK, Ten S, Hwa V, Rosenfeld RG** 2011 A novel Y332C missense mutation in the intracellular domain of the human growth hormone receptor (GHR) does not alter STAT5b signaling: redundancy of GHR in-

tracellular tyrosines involved in STAT5b signaling. Horm Res Paediatr 75:187–199
28. **Milward A, Metherell L, Maamra M, Barahona MJ, Wilkinson IR, Camacho-Hübner C, Savage MO, Bidlingmaier M, Bidlingmaier CM, Clark AJ, Ross RJ, Webb SM** 2004 Growth hormone insensitivity syndrome due to a growth hormone receptor truncated after box 1 resulting in isolated failure of STAT 5 signal transduction. J Clin Endocrinol Metab 89:1259–1266
29. **Jin H, Lanning NJ, Carter-Su C** 2008 JAK2, but not Src family kinases, is required for STAT, ERK, and Akt signaling in response to growth hormone in preadipocytes and hepatoma cells. Mol Endocrinol 22:1825–1841
30. **Rowlinson SW, Yoshizato H, Barclay JL, Brooks AJ, Behncken SN, Kerr LM, Millard K, Palethorpe K, Nielsen K, Clyde-Smith J, Hancock JF, Waters MJ** 2008 An agonist-induced conformational change in the growth hormone receptor determines the choice of signalling pathway. Nat Cell Biol 10:740–747
31. **Barclay JL, Kerr LM, Arthur L, Rowland JE, Nelson CN, Ishikawa M, d'Aniello EM, White M, Noakes PG, Waters MJ** 2010 In vivo targeting of the growth hormone receptor (GHR) box1 sequence demonstrates that the GHR does not signal exclusively through JAK2. Mol Endocrinol 24: 204–217
32. **Chia DJ, Varco-Merth B, Rotwein P** 2010 Dispersed chromosomal Stat5b-binding elements mediate growth hormone-activated insulin-like growth factor-I gene transcription. J Biol Chem 285:17636–17647
33. **Wang Y, Jiang H** 2005 Identification of a distal STAT-binding DNA region that may mediate growth hormone regulation of insulin-like growth factor-I gene expression. J Biol Chem 280:10955–10963
34. **Adams TE, Epa VC, Garrett TP, Ward CW** 2000 Structure and function of the type 1 insulin-like growth factor receptor. Cell Mol Life Sci 57:1050–1093
35. **Fowden AL** 2003 The insulin-like growth factors and feto-placental growth. Placenta 24:803–812
36. **Lupu F, Terwilliger JD, Lee K, Segre GV, Efstratiadis A** 2001 Roles of growth hormone and insulin-like growth factor-1 in mouse postnatal growth. Dev Biol 229:141–162
37. **Woods KA, Camacho-Hübner C, Savage MO, Clark AJ** 1996 Intrauterine growth retardation and post-natal growth failure associated with deletion of the insulin-like growth factor-I gene. N Engl J Med 335:1363–1367
38. **Netchine I, Azzi S, Le Bouc Y, Savage MO** 2011 IGF1 molecular anomalies demonstrate its critical role in fetal, postnatal growth and brain development. Best Pract Res Clin Endocrinol Metab 25:181–190
39. **Walenkamp MJ, van der Kamp HJ, Pereira AM, Kant SG, van Duyvenvoorde HA, Kruithof MF, Breuning MH, Romijn JA, Karperien M, Wit JM** 2006 A variable degree of intrauterine and postnatal growth retardation in a family with a missense mutation in the insulin-like growth factor I receptor. J Clin Endocrinol Metab 91:3062–3470
40. **Ballesteros M, Leung KC, Ross RJ, Iismaa TP, Ho KK** 2000 Distribution and abundance of messenger ribonucleic acid for growth hormone receptor isoforms in human tissues. J Clin Endocrinol Metab 85:2865–2871
41. **Brown RJ, Adams JJ, Pelekanos RA, Wan Y, McKinstry WJ, Palethorpe K, Seeber RM, Monks TA, Eidne KA, Parker MW, Waters MJ** 2005 Model for growth hormone receptor activation based on subunit rotation within a receptor dimer. Nat Struct Mol Biol 12:814–821
42. **Brooks AJ, Waters MJ** 2010 The growth hormone receptor: mechanism of activation and clinical implications. Nat Rev Endocrinol 6:515–525
43. **Argetsinger LS, Campbell GS, Yang X, Witthuhn BA, Silvennoinen O, Ihle JN, Carter-Su C** 1993 Identification of JAK2 as a growth hormone receptor-associated kinase. Cell 74:237–244
44. **Diniz ET, Jorge AA, Arnhold IJ, Rosenbloom AL, Bandeira F** 2008 Novel missense mutation (pY113X) in the human growth hormone receptor gene in a Brazilian patient with Laron syndrome. [in Portuguese] Arq Bras Endocrinol Metabol 52:1264–1271
45. **Baumann G** 2002 Genetic characterization of growth hormone deficiency and resistance: implications for treatment with recombinant growth hormone. Am J Pharmacogenomics 2:93–111
46. **Metherell LA, Akker SA, Munroe PB, Rose SJ, Caulfield M, Savage MO, Chew SL, Clark AJ** 2001 Pseudoexon activation as a novel mechanism for disease resulting in atypical growth hormone insensitivity. Am J Hum Genet 69: 641–646
47. **Maamra M, Milward A, Esfahani HZ, Abbott LP, Metherell LA, Savage MO, Clark AJ, Ross RJ** 2006 A 36 residues insertion in the dimerization domain of the growth hormone receptor results in defective trafficking rather than impaired signalling. J Endocrinol 188:251–261
48. **Akker SA, Misra S, Aslam S, Morgan EL, Smith PJ, Khoo B, Chew SL** 2007 Pre-spliceosomal binding of U1 small nuclear ribonucleoprotein (RNP) and heterogenous nuclear RNP E1 is associated with suppression of a growth hormone receptor pseudoexon. Mol Endocrinol 21:2529–2540
49. **David A, Camacho-Hübner C, Bhangoo A, Rose SJ, Miraki-Moud F, Akker SA, Butler GE, Ten S, Clayton PE, Clark AJ, Savage MO, Metherell LA** 2007 An intronic growth hormone receptor mutation causing activation of a pseudoexon is associated with a broad spectrum of growth hormone insensitivity phenotypes. J Clin Endocrinol Metab 92:655–659
50. **Ayling RM, Ross R, Towner P, Von Laue S, Finidori J, Moutoussamy S, Buchanan CR, Clayton PE, Norman MR** 1997 A dominant-negative mutation of the growth hormone receptor causes familial short stature. Nat Genet 16: 13–14
51. **Iida K, Takahashi Y, Kaji H, Nose O, Okimura Y, Abe H, Chihara K** 1998 Growth hormone (GH) insensitivity syndrome with high serum GH-binding protein levels caused by a heterozygous splice site mutation of the GH receptor gene producing a lack of intracellular domain. J Clin Endocrinol Metab 83:531–537
52. **Fang P, Riedl S, Amselem S, Pratt KL, Little BM, Haeusler G, Hwa V, Frisch H, Rosenfeld RG** 2007 Primary growth hormone (GH) insensitivity and insulin-like growth factor deficiency caused by novel compound heterozygous mutations of the GH receptor gene: genetic and functional studies of simple and compound heterozygous states. J Clin Endocrinol Metab 92:2223–2231
53. **Aisenberg J, Auyeung V, Pedro HF, Sugalski R, Chartoff A, Rothenberg R, Derr MA, Hwa V, Rosenfeld RG** 2010 Atypical GH insensitivity syndrome and severe insulin-

like growth factor-I deficiency resulting from compound heterozygous mutations of the GH receptor, including a novel frameshift mutation affecting the intracellular domain. Horm Res Paediatr 74:406–411

54. **Aalbers AM, Chin D, Pratt KL, Little BM, Frank SJ, Hwa V, Rosenfeld RG** 2009 Extreme elevation of serum growth hormone-binding protein concentrations resulting from a novel heterozygous splice site mutation of the growth hormone receptor gene. Horm Res 71:276–284
55. **Rosenfeld RG, Belgorosky A, Camacho-Hubner C, Savage MO, Wit JM, Hwa V** 2007 Defects in growth hormone receptor signaling. Trends Endocrinol Metab 18:134–141
56. **Kofoed EM, Hwa V, Little B, Woods KA, Buckway CK, Tsubaki J, Pratt KL, Bezrodnik L, Jasper H, Tepper A, Heinrich JJ, Rosenfeld RG** 2003 Growth-hormone insensitivity associated with a STAT5b mutation. N Engl J Med 349:1139–1147
57. **Hwa V, Camacho-Hübner C, Little BM, David A, Metherell LA, El-Khatib N, Savage MO, Rosenfeld RG** 2007 Growth hormone insensitivity and severe short stature in siblings: a novel mutation at the exon 13-intron 13 junction of the STAT5b gene. Horm Res 68:218–224
58. **Bernasconi A, Marino R, Ribas A, Rossi J, Ciaccio M, Oleastro M, Ornani A, Paz R, Rivarola MA, Zelazko M, Belgorosky A** 2006 Characterization of immunodeficiency in a patient with growth hormone insensitivity secondary to a novel STAT5b gene mutation. Pediatrics 118:e1584–e1592
59. **Hwa V, Little B, Adiyaman P, Kofoed EM, Pratt KL, Ocal G, Berberoglu M, Rosenfeld RG** 2005 Severe growth hormone insensitivity resulting from total absence of signal transducer and activator of transcription 5b. J Clin Endocrinol Metab 90:4260–4266
60. **Vidarsdottir S, Walenkamp MJ, Pereira AM, Karperien M, van Doorn J, van Duyvenvoorde HA, White S, Breuning MH, Roelfsema F, Kruithof MF, van Dissel J, Janssen R, Wit JM, Romijn JA** 2006 Clinical and biochemical characteristics of a male patient with a novel homozygous STAT5b mutation. J Clin Endocrinol Metab 91:3482–3485
61. **Rubinstein A, Bernstein LJ, Charytan M, Krieger BZ, Ziprkowski M** 1988 Corticosteroid treatment for pulmonary lymphoid hyperplasia in children with the acquired immunodeficiency syndrome. Pediatr Pulmonol 4:13–17
62. **Hwa V, Little B, Kofoed EM, Rosenfeld RG** 2004 Transcriptional regulation of insulin-like growth factor-I by interferon-γ requires STAT-5b. J Biol Chem 279:2728–2736
63. **Fang P, Kofoed EM, Little BM, Wang X, Ross RJ, Frank SJ, Hwa V, Rosenfeld RG** 2006 A mutant signal transducer and activator of transcription 5b, associated with growth hormone insensitivity and insulin-like growth factor-I deficiency, cannot function as a signal transducer or transcription factor. J Clin Endocrinol Metab 91:1526–1534
64. **Cohen AC, Nadeau KC, Tu W, Hwa V, Dionis K, Bezrodnik L, Teper A, Gaillard M, Heinrich J, Krensky AM, Rosenfeld RG, Lewis DB** 2006 Cutting edge: decreased accumulation and regulatory function of CD4+CD25high T cells in human STAT5b deficiency. J Immunol 177:2770–2774
65. **Chen X, Vinkemeier U, Zhao Y, Jeruzalmi D, Darnell Jr JE, Kuriyan J** 1998 Crystal structure of a tyrosine phosphorylated STAT-1 dimer bound to DNA. Cell 93:827–839
66. **Chia DJ, Subbian E, Buck TM, Hwa V, Rosenfeld RG, Skach WR, Shinde U, Rotwein P** 2006 Aberrant folding of a mutant STAT5b causes growth hormone insensitivity and proteasomal dysfunction. J Biol Chem 281:6552–6558
67. **Dupuis S, Jouanguy E, Al-Hajjar S, Fieschi C, Al-Mohsen IZ, Al-Jumaah S, Yang K, Chapgier A, Eidenschenk C, Eid P, Al Ghonaium A, Tufenkeji H, Frayha H, Al-Gazlan S, Al-Rayes H, Schreiber RD, Gresser I, Casanova JL** 2003 Impaired response to interferon-α/β and lethal viral disease in human STAT1 deficiency. Nat Genet 33:388–391
68. **Lappalainen I, Thusberg J, Shen B, Vihinen M** 2008 Genome wide analysis of pathogenic SH2 domain mutations. Proteins 72:779–792
69. **Pugliese-Pires PN, Tonelli CA, Dora JM, Silva PC, Czepielewski M, Simoni G, Arnhold IJ, Jorge AA** 2010 A novel STAT5B mutation causing GH insensitivity syndrome associated with hyperprolactinemia and immune dysfunction in two male siblings. Eur J Endocrinol 163:349–355
70. **Martinez A, Scaglia PA, Rivas M, Bezrodnik L, Gaillard MI, Heinrich JJ, Ballerini MG, Jasper HG, Domene HM** 2007 Novel STAT5b gene mutation in a patient presenting GH insensitivity and immunodeficiency. Horm Res 68(Suppl 1):33
71. **Boyanovsky A, Lozano A, Testa G, Munoz L, Marino R, Bernasconi A, Belgorosky A, Miras M,** Growth hormone insensitivity and immunodeficiency: mutation in the STAT5B gene. Proc 8th Joint Meeting of the Lawson Wilkins Pediatric Endocrine Society/European Society for Paediatric Endocrinology, New York, 2009 (Abstract P01-067)
72. **Tartaglia M, Mehler EL, Goldberg R, Zampino G, Brunner HG, Kremer H, van der Burgt I, Crosby AH, Ion A, Jeffery S, Kalidas K, Patton MA, Kucherlapati RS, Gelb BD** 2001 Mutations in *PTPN11*, encoding the protein tyrosine phosphatase SHP-2, cause Noonan syndrome. Nat Genet 29:465–468
73. **Brasil AS, Pereira AC, Wanderley LT, Kim CA, Malaquias AC, Jorge AA, Krieger JE, Bertola DR** 2010 PTPN11 and KRAS gene analysis in patients with Noonan and Noonan-like syndromes. Genet Test Mol Biomarkers 14:425–432
74. **Kim SO, Jiang J, Yi W, Feng GS, Frank SJ** 1998 Involvement of the Src homology 2-containing tyrosine phosphatase SHP2 in growth hormone signaling. J Biol Chem 273:2344–2354
75. **Stofega MR, Herrington J, Billestrup N, Carter-Su C** 2000 Mutation of the SHP2 binding site in growth hormone (GH) receptor prolongs GH-promoted tyrosyl phosphorylation of GH receptor, JAK2, and STAT5B. Mol Endocrinol 14:1338–1350
76. **Binder G, Neuer K, Ranke MB, Wittekindt NE** 2005 PTPN11 mutations are associated with mild growth hormone resistance in individuals with Noonan syndrome. J Clin Endocrinol Metab 90:5377–5381
77. **Limal JM, Parfait B, Cabrol S, Bonnet D, Leheup B, Lyonnet S, Vidaud M, Le Bouc Y** 2006 Noonan syndrome: relationships between genotype, growth, and growth factors. J Clin Endocrinol Metab 91:300–306
78. **Binder G** 2009 Response to growth hormone in short children with Noonan syndrome: correlation to genotype. Horm Res 72(Suppl 2):52–56

79. **Woods KA, Camacho-Hübner C, Bergman RN, Barter D, Clark AJ, Savage MO** 2000 Effects of insulin-like growth factor I (IGF-I) therapy on body composition and insulin resistance in IGF-I gene deletion. J Clin Endocrinol Metab 85:1407–1411
80. **Camacho-Hübner C, Woods KA, Miraki-Moud F, Hindmarsh PC, Clark AJ, Hansson Y, Johnston A, Baxter RC, Savage MO** 1999 Effects of recombinant human insulin-like growth factor-I (IGF-I) therapy on the growth hormone-IGF system of a patient with partial IGF-I gene deletion. J Clin Endocrinol Metab 84:1611–1616
81. **Bonapace G, Concolino D, Formicola S, Strisciuglio P** 2003 A novel mutation in a patient with insulin-like growth factor 1 (IGF1) deficiency. J Med Genet 40:913–917
82. **Walenkamp MJ, Karperien M, Pereira AM, Hilhorst-Hofstee Y, van Doorn J, Chen JW, Mohan S, Denley A, Forbes B, van Duyvenvoorde HA, van Thiel SW, Sluimers CA, Bax JJ, de Laat JA, Breuning MB, Romijn JA, Wit JM** 2005 Homozygous and heterozygous expression of a novel insulin-like growth factor-I mutation. J Clin Endocrinol Metab 90:2855–2864
83. **Netchine I, Azzi S, Houang M, Seurin D, Perin L, Ricort JM, Daubas C, Legay C, Mester J, Herich R, Godeau F, Le Bouc Y** 2009 Partial primary deficiency of insulin-like growth factor (IGF)-I activity associated with IGF-1 mutation demonstrates its critical role in growth and brain development. J Clin Endocrinol Metab 94:3913–3921
84. **Netchine I, Rossignol S, Dufourg MN, Azzi S, Rousseau A, Perin L, Houang M, Steunou V, Esteva B, Thibaud N, Demay MC, Danton F, Petriczko E, Bertrand AM, Heinrichs C, Carel JC, Loeuille GA, Pinto G, Jacquemont ML, Gicquel C, Cabrol S, Le Bouc Y** 2007 11p15 imprinting center region 1 loss of methylation is a common and specific cause of typical Russell-Silver syndrome: clinical scoring system and epigenetic-phenotypic correlations. J Clin Endocrinol Metab 92:3148–3154
85. **van Gemund JJ, Laurent de Angulo MS, van Gelderen HH** 1970 Familial prenatal dwarfism with elevated serum immuno-reactive growth hormone levels and end-organ unresponsiveness. Maandschr Kindergeneeskd 37:372–382
86. **Coutinho DC, Coletta RR, Costa EM, Pachi PR, Boguszewski MC, Damiani D, Mendonca BB, Arnhold IJ, Jorge AA** 2007 Polymorphisms identified in the upstream core polyadenylation signal of IGF1gene exon 6 do not cause pre- and postnatal growth impairment. J Clin Endocrinol Metab 92:4889–4892
87. **van Duyvenvoorde HA, van Setten PA, Walenkamp MJ, van Doorn J, Koenig J, Gauguin L, Oostdijk W, Ruivenkamp CA, Losekoot M, Wade JD, De Meyts P, Karperien M, Noordam C, Wit JM** 2010 Short stature associated with a novel heterozygous mutation in the insulin-like growth factor 1 gene. J Clin Endocrinol Metab 95:E363–E367
88. **van Duyvenvoorde HA, van Doorn J, Koenig J, Gauguin L, Oostdijk W, Wade JD, Karperien M, Ruivenkamp CA, Losekoot M, van Setten PA, Walenkamp MJ, Noordam C, De Meyts P, Wit JM** 2011 The severe short stature in two siblings with a heterozygous *IGF1* mutation is not caused by a dominant negative effect of the putative truncated protein. Growth Horm IGF Res 21:44–50
89. **Fuqua JS, Hwa V, Rosenfeld RG,** Identification of a heterozygous IGF-1 gene splice mutation. Proc 8th Joint Meeting of the Lawson Wilkins Pediatric Endocrine Society/European Society for Paediatric Endocrinology, New York, 2009 (Abstract P01-075)
90. **Leong SR, Baxter RC, Camerato T, Dai J, Wood WI** 1992 Structure and functional expression of the acid-labile subunit of the insulin-like growth factor-binding protein complex. Mol Endocrinol 6:870–876
91. **Ooi GT, Cohen FJ, Tseng LY, Rechler MM, Boisclair YR** 1997 Growth hormone stimulates transcription of the gene encoding the acid-labile subunit (ALS) of the circulating insulin-like growth factor-binding protein complex and ALS promoter activity in rat liver. Mol Endocrinol 11:997–1007
92. **Baxter RC** 1994 Insulin-like growth factor binding proteins in the human circulation: a review. Horm Res 42: 140–144
93. **Boisclair YR, Rhoads RP, Ueki I, Wang J, Ooi GT** 2001 The acid-labile subunit (ALS) of the 150 kDa IGF-binding protein complex: an important but forgotten component of the circulating IGF system. J Endocrinol 170:63–70
94. **Domené HM, Scaglia PA, Lteif A, Mahmud FH, Kirmani S, Frystyk J, Bedecarrás P, Gutiérrez M, Jasper HG** 2007 Phenotypic effects of null and haploinsufficiency of acid-labile subunit in a family with two novel IGFALS gene mutations. J Clin Endocrinol Metab 92:4444–4450
95. **Domené HM, Hwa V, Argente J, Wit JM, Wit JM, Camacho-Hübner C, Jasper HG, Pozo J, van Duyvenvoorde HA, Yakar S, Fofanova-Gambetti OV, Rosenfeld RG** 2009 Human acid-labile subunit deficiency: clinical, endocrine and metabolic consequences. Horm Res 72:129–141
96. **David A, Rose SJ, Miraki-Moud F, Metherell LA, Savage MO, Clark AJ, Camacho-Hübner C** 2010 Acid-labile subunit deficiency and growth failure: description of two novel cases. Horm Res Paediatr 73:328–334
97. **Heath KE, Argente J, Barrios V, Pozo J, Díaz-González F, Martos-Moreno GA, Caimari M, Gracia R, Campos-Barros A** 2008 Primary acid-labile subunit deficiency due to recessive IGFALS mutations results in postnatal growth deficit associated with low circulating insulin growth factor (IGF)-I, IGF binding protein-3 levels, and hyperinsulinemia. J Clin Endocrinol Metab 93:1616–1624
98. **Fofanova-Gambetti OV, Hwa V, Wit JM, Domene HM, Argente J, Bang P, Högler W, Kirsch S, Pihoker C, Chiu HK, Cohen L, Jacobsen C, Jasper HG, Haeusler G, Campos-Barros A, Gallego-Gómez E, Gracia-Bouthelier R, van Duyvenvoorde HA, Pozo J, Rosenfeld RG** 2010 Impact of heterozygosity for acid-labile subunit (IGFALS) gene mutations on stature: results from the International Acid-Labile Subunit Consortium. J Clin Endocrinol Metab 95:4184–4191
99. **Abuzzahab MJ, Schneider A, Goddard A, Grigorescu F, Lautier C, Keller E, Kiess W, Klammt J, Kratzsch J, Osgood D, Pfäffle R, Raile K, Seidel B, Smith RJ, Chernausek SD** 2003 IGF-I receptor mutations resulting in intrauterine and postnatal growth retardation. N Engl J Med 349: 2211–2222
100. **Kawashima Y, Kanzaki S, Yang F, Kinoshita T, Hanaki K, Nagaishi J, Ohtsuka Y, Hisatome I, Ninomoya H, Nanba E, Fukushima T, Takahashi S** 2005 Mutation at cleavage site of insulin-like growth factor receptor in a short-stature

child born with intrauterine growth retardation. J Clin Endocrinol Metab 90:4679–4687

101. **Fang P, Schwartz ID, Johnson BD, Derr MA, Roberts Jr CT, Hwa V, Rosenfeld RG** 2009 Familial short stature caused by haploinsufficiency of the insulin-like growth factor I receptor due to nonsense-mediated messenger ribonucleic acid decay. J Clin Endocrinol Metab 94:1740–1747
102. **Inagaki K, Tiulpakov A, Rubtsov P, Sverdlova P, Peterkova V, Yakar S, Terekhov S, LeRoith D** 2007 A familial insulin-like growth factor-I receptor mutant leads to short stature: clinical and biochemical characterization. J Clin Endocrinol Metab 92:1542–1548
103. **Wallborn T, Wüller S, Klammt J, Kruis T, Kratzsch J, Schmidt G, Schlicke M, Müller E, van de Leur HS, Kiess W, Pfäffle R** 2010 A heterozygous mutation of the insulin-like growth factor-I receptor causes retention of the nascent protein in the endoplasmic reticulum and results in intrauterine and postnatal growth retardation. J Clin Endocrinol Metab 95:2316–2324
104. **Kruis T, Klammt J, Galli-Tsinopoulou A, Wallborn T, Schlicke M, Müller E, Kratzsch J, Körner A, Odeh R, Kiess W, Pfäffle R** 2010 Heterozygous mutation within a kinase-conserved motif of the insulin-like growth factor I receptor causes intrauterine and postnatal growth retardation. J Clin Endocrinol Metab 95:1137–1142
105. **Ester WA, van Duyvenvoorde HA, de Wit CC, Broekman AJ, Ruivenkamp CA, Govaerts LC, Wit JM, Hokken-Koelega AC, Losekoot M** 2009 Two short children born small for gestational age with insulin-like growth factor 1 receptor haploinsufficiency illustrate the heterogeneity of its phenotype. J Clin Endocrinol Metab 94:4717–4727
106. **Kowarski AA, Schneider J, Ben-Galim E, Weldon VV, Daughaday WH** 1978 Growth failure with normal serum RIA-GH and low somatomedin activity: somatomedin restoration and growth acceleration after exogenous GH. J Clin Endocrinol Metab 47:461–464
107. **Takahashi Y, Kaji H, Okimura Y, Goji K, Abe H, Chihara K** 1996 Short stature caused by a mutant growth hormone. N Engl J Med 334:432–436
108. **Takahashi Y, Shirono H, Arisaka O, Takahashi K, Yagi T, Koga J, Kaji H, Okimura Y, Abe H, Tanaka T, Chihara K** 1997 Biologically inactive growth hormone caused by an amino acid substitution. J Clin Invest 100:1159–1165
109. **Besson A, Salemi S, Deladoëy J, Vuissoz JM, Eblé A, Bidlingmaier M, Bürgi S, Honegger U, Flück C, Mullis PE** 2005 Short stature caused by a biologically inactive mutant growth hormone (GH-C53S). J Clin Endocrinol Metab 90:2493–2499
110. **Cogan JD, Phillips 3rd JA** 2006 GH1 gene deletions and IGHD type 1A. Pediatr Endocrinol Rev 3:480–488
111. **Phillips 3rd JA, Hjelle BL, Seeburg PH, Zachmann M** 1981 Molecular basis for familial isolated growth hormone deficiency. Proc Natl Acad Sci USA 78:6372–6375
112. **Procter AM, Phillips 3rd JA, Cooper DN** 1998 The molecular genetics of growth hormone deficiency. Hum Genet 103:255–272
113. **Alatzoglou KS, Dattani MT** 2010 Genetic causes and treatment of isolated growth hormone deficiency—an update. Nat Rev Endocrinol 6:562–576
114. **Riedl S, Frisch H** 2006 Effects of growth hormone (GH) and insulin-like growth factor-I therapy in patients with gene defects in the GH axis. J Pediatr Endocrinol Metab 19:229–236
115. **Speiser PW, White PC** 2003 Congenital adrenal hyperplasia. N Engl J Med 349:776–788
116. **Woods KA, Dastot F, Preece MA, Clark AJ, Postel-Vinay MC, Chatelain PG, Ranke MB, Rosenfeld RG, Amselem S, Savage MO** 1997 Phenotype: genotype relationships in growth hormone insensitivity syndrome. J Clin Endocrinol Metab 82:3529–3535
117. **Burren CP, Woods KA, Rose SJ, Tauber M, Price DA, Heinrich U, Gilli G, Razzaghy-Azar M, Al-Ashwal A, Crock PA, Rochiccioli P, Yordam N, Ranke MB, Chatelain PG, Preece MA, Rosenfeld RG, Savage MO** 2001 Clinical and endocrine characteristics in atypical and classical growth hormone insensitivity syndrome. Horm Res 55:125–130
118. **Guevara-Aguirre J, Rosenbloom AL, Fielder PJ, Diamond Jr FB, Rosenfeld RG** 1993 Growth hormone receptor deficiency in Ecuador: clinical and biochemical phenotype in two populations. J Clin Endocrinol Metab 76:417–423
119. **Blum WF, Cotterill AM, Postel-Vinay MC, Ranke MB, Savage MO, Wilton P** 1994 Improvement of diagnostic criteria in growth hormone insensitivity syndrome: solutions and pitfalls. Pharmacia Study Group on Insulin-like Growth Factor I Treatment in Growth Hormone Insensitivity Syndromes. Acta Paediatr Suppl 399:117–124
120. **Cotterill AM, Holly JM, Taylor AM, Davies SC, Coulson VJ, Preece MA, Wass JA, Savage MO** 1992 The insulin-like growth factor (IGF)-binding proteins and IGF bioactivity in Laron-type dwarfism. J Clin Endocrinol Metab 74:56–63
121. **Burren CP, Wanek D, Mohan S, Cohen P, Guevara-Aguirre J, Rosenfeld RG** 1999 Serum levels of insulin-like growth factor binding proteins in Ecuadorean children with growth hormone insensitivity. Acta Paediatr Suppl 88:185–191; discussion 192
122. **Zhu Q, Watanabe C, Liu T, Hollenbaugh D, Blaese RM, Kanner SB, Aruffo A, Ochs HD** 1997 Wiskott-Aldrich syndrome/X-linked thrombocytopenia: WASP gene mutations, protein expression, and phenotype. Blood 90:2680–2689
123. **Lemahieu V, Gastier JM, Francke U** 1999 Novel mutations in the Wiskott-Aldrich syndrome protein gene and their effects on transcriptional, translational, and clinical phenotypes. Hum Mutat 14:54–66
124. **Savage MO, Burren CP, Rosenfeld RG** 2010 The continuum of growth hormone-IGF-I axis defects causing short stature: diagnostic and therapeutic challenges. Clin Endocrinol (Oxf) 72:721–728
125. **Cohen P, Rogol AD, Deal CL, Saenger P, Reiter EO, Ross JL, Chernausek SD, Savage MO, Wit JM, on behalf of the 2007 ISS Consensus Workshop participants** 2008 Consensus statement on the diagnosis and treatment of children with idiopathic short stature: a summary of the Growth Hormone Research Society, the Lawson Wilkins Pediatric Endocrine Society, and the European Society for Paediatric Endocrinology Workshop. J Clin Endocrinol Metab 93:4210–4217
126. **Growth Hormone Research Society** 2000 Consensus guidelines for the diagnosis and treatment of growth hormone (GH) deficiency in childhood and adolescence: Sum-

mary Statement of the GH Research Society. J Clin Endocrinol Metab 85:3990–3993
127. **Cianfarani S, Liguori A, Boemi S, Maghnie M, Iughetti L, Wasniewska M, Street ME, Zucchini S, Aimaretti G, Germani D** 2005 Inaccuracy of insulin-like growth factor (IGF) binding protein (IGFBP-3) assessment in the diagnosis of growth hormone (GH) deficiency from childhood to young adulthood: association to low GH dependency of IGF-II and presence of circulating IGFBP-3 18-kilodalton fragment. J Clin Endocrinol Metab 90:6028–6034
128. **Juul A, Bang P, Hertel NT, Main K, Dalgaard P, Jørgensen K, Müller J, Hall K, Skakkebaek NE** 1994 Serum insulin-like growth factor-I in 1030 healthy children, adolescents and adults: relation to age, sex, stage of puberty, testicular size and body mass index. J Clin Endocrinol Metab 78: 744–752
129. **Juul A, Dalgaard P, Blum WF, Bang P, Hall K, Michaelsen KF, Müller J, Skakkebaek NE** 1995 Serum levels of insulin-like growth factor (IGF)- binding protein-3 (IGFBP-3) in healthy infants, children and adolescents: the relation to IGF-I, IGF-II, IGFBP-1, IGFBP-2, age, sex, body mass index and pubertal maturation. J Clin Endocrinol Metab 80:2534–2542
130. **Rosenfeld RG, Kemp SF, Hintz RL** 1981 Constancy of somatomedin response to growth hormone treatment of hypopituitary dwarfism, and lack of correlation with growth rate. J Clin Endocrinol Metab 53:611–617
131. **Rosenfeld RG** 2005 The IGF system: new developments relevant to pediatric practice. In: Cianfarani S, Clemmons DR, Savage MO, eds. IGF-I and IGF binding proteins: basic research and clinical management. Endocrine Development, vol 9. Basel, Switzerland: Karger; 1–10
132. **Selva KA, Buckway CK, Sexton G, Pratt KL, Tjoeng E, Guevara-Aguirre J, Rosenfeld RG** 2003 Reproducibility in patterns of IGF generation tests with special reference to idiopathic short stature. Horm Res 60:237–246
133. **Midyett LK, Rogol AD, Van Meter QL, Frane J, Bright GM; MS301 Study Group** 2010 Recombinant insulin-like growth factor (IGF)-I treatment in short children with low IGF-I levels: first-year results from a randomized clinical trial. J Clin Endocrinol Metab 95:611–619
134. **Buckway CK, Guevara-Aguirre J, Pratt KL, Burren CP, Rosenfeld RG** 2001 The IGF-I generation test revisited: a marker of GH sensitivity. J Clin Endocrinol Metab 86: 5176–5183
135. **Jorge AA, Souza SC, Arnhold IJ, Mendonca BB** 2002 Poor reproducibility of the IGF-I and IGF binding protein-3 generation test in children with short stature and normal coding region of the GH receptor gene. J Clin Endocrinol Metab 87:469–472
136. **Shevah O, Borrelli P, Rubinstein M, Laron Z** 2003 Identification of two novel mutations in the human growth hormone receptor gene. J Endocrinol Invest 26:604–608
137. **Pantel J, Grulich-Henn J, Bettendorf M, Strasburger CJ, Heinrich U, Amselem S** 2003 Heterozygous nonsense mutation in exon 3 of the growth hormone receptor (GHR) in severe GH insensitivity (Laron syndrome) and the issue of the origin and function of the GHRd3 isoform. J Clin Endocrinol Metab 88:1705–1710
138. **Amselem S, Sobrier ML, Duquesnoy P, Rappaport R, Postel-Vinay MC, Gourmelen M, Dallapiccola B, Goossens M** 1991 Recurrent nonsense mutations in the growth hormone receptor from patients with Laron dwarfism. J Clin Invest 87:1098–1102
139. **Amselem S, Duquesnoy P, Duriez B, Dastot F, Sobrier ML, Valleix S, Goossens M** 1993 Spectrum of growth hormone receptor mutations and associated haplotypes in Laron syndrome. Hum Mol Genet 2:355–359
140. **Berg MA, Argente J, Chernausek S, Gracia R, Guevara-Aguirre J, Hopp M, Pérez-Jurado L, Rosenbloom A, Toledo SP, Francke U** 1993 Diverse growth hormone receptor gene mutations in Laron syndrome. Am J Hum Genet 52: 998–1005
141. **Putzolu M, Meloni A, Loche S, Pischedda C, Cao A, Moi P** 1997 A homozygous nonsense mutation of the human growth hormone receptor gene in a Sardinian boy with Laron-type dwarfism. J Endocrinol Invest 20:286–288
142. **Sobrier ML, Dastot F, Duquesnoy P, Kandemir N, Yordam N, Goossens M, Amselem S** 1997 Nine novel growth hormone receptor gene mutations in patients with Laron syndrome. J Clin Endocrinol Metab 82:435–437
143. **Tiulpakov A, Rubtsov P, Dedov I, Peterkova V, Bezlepkina O, Chrousos GP, Hochberg Z** 2005 A novel C-terminal growth hormone receptor (GHR) mutation results in impaired GHR-STAT5 but normal STAT-3 signaling. J Clin Endocrinol Metab 90:542–547
144. **Goddard AD, Covello R, Luoh SM, Clackson T, Attie KM, Gesundheit N, Rundle AC, Wells JA, Carlsson LM** 1995 Mutations of the growth hormone receptor in children with idiopathic short stature. The Growth Hormone Insensitivity Study Group. N Engl J Med 333:1093–1098
145. **Fassone L, Corneli G, Bellone S, Camacho-Hübner C, Aimaretti G, Cappa M, Ubertini G, Bona G** 2007 Growth hormone receptor gene mutations in two Italian patients with Laron syndrome. J Endocrinol Invest 30:417–420
146. **Fang P, Girgis R, Little BM, Pratt KL, Guevara-Aguirre J, Hwa V, Rosenfeld RG** 2008 Growth hormone (GH) insensitivity and insulin-like growth factor-I deficiency in Inuit subjects and an Ecuadorian cohort: functional studies of two codon 180 GH receptor gene mutations. J Clin Endocrinol Metab 93:1030–1037
147. **Berg MA, Peoples R, Pérez-Jurado L, Guevara-Aguirre J, Rosenbloom AL, Laron Z, Milner RD, Francke U** 1994 Receptor mutations and haplotypes in growth hormone receptor deficiency: a global survey and identification of the Ecuadorean E180splice mutation in an oriental Jewish patient. Acta Paediatr Suppl 399:112–114
148. **Kaji H, Nose O, Tajiri H, Takahashi Y, Iida K, Takahashi T, Okimura Y, Abe H, Chihara K** 1997 Novel compound heterozygous mutations of growth hormone (GH) receptor gene in a patient with GH insensitivity syndrome. J Clin Endocrinol Metab 82:3705–3709
149. **Quinteiro C, Castro-Feijoo L, Loidi L, Barreiro J, de la Fuente M, Dominguez F, Pombo M** 2002 Novel mutation involving the translation initiation codon of the growth hormone receptor gene (GHR) in a patient with Laron syndrome. J Pediatr Endocrinol Metab 15:1041–1045
150. **Chen X, Song F, Dai Y, Bao X, Jin Y** 2003 A novel mutation of the growth hormone receptor gene (GHR) in a Chinese girl with Laron syndrome. J Pediatr Endocrinol Metab 16:1183–1189
151. **Ying YQ, Wei H, Cao LZ, Lu JJ, Luo XP** 2007 Clinical features and growth hormone receptor gene mutations of

patients with Laron syndrome from a Chinese family. Zhongguo Dang Dai Er Ke Za Zhi 9:335–338
152. **Shevah O, Rubinstein M, Laron Z** 2004 Molecular defects of the growth hormone receptor gene, including a new mutation, in Laron syndrome patients in Israel: relationship between defects and ethnic groups. Isr Med Assoc J 6:630–633
153. **Arman A, Yüksel B, Coker A, Sarioz O, Temiz F, Topaloglu AK** 2010 Novel growth hormone receptor gene mutation in a patient with Laron syndrome. J Pediatr Endocrinol Metab 23:407–414
154. **Walker JL, Crock PA, Behncken SN, Rowlinson SW, Nicholson LM, Boulton TJ, Waters MJ** 1998 A novel mutation affecting the interdomain link region of the growth hormone receptor in a Vietnamese girl, and response to long-term treatment with recombinant human insulin-like growth factor-I and luteinizing hormone-releasing hormone analogue. J Clin Endocrinol Metab 83:2554–2561
155. **Sanchez JE, Perera E, Baumbach L, Cleveland WW** 1998 Growth hormone receptor mutations in children with idiopathic short stature. J Clin Endocrinol Metab 83:4079–4083
156. **Duquesnoy P, Sobrier ML, Duriez B, Dastot F, Buchanan CR, Savage MO, Preece MA, Craescu CT, Blouquit Y, Goossens M** 1994 A single amino acid substitution in the exoplasmic domain of the human growth hormone (GH) receptor confers familial GH resistance (Laron syndrome) with positive GH-binding activity by abolishing receptor homodimerization. EMBO J 13:1386–1395
157. **Yang C, Chen JY, Lai CC, Lin HC, Yeh GC, Hsu HH** 2004 Clinical, biochemical and molecular investigations of three Taiwanese children with Laron syndrome. J Pediatr Endocrinol Metab 17:165–171
158. **Wojcik J, Berg MA, Esposito N, Geffner ME, Sakati N, Reiter EO, Dower S, Francke U, Postel-Vinay MC, Finidori J** 1998 Four contiguous amino acid substitutions, identified in patients with Laron syndrome, differently affect the binding affinity and intracellular trafficking of the growth hormone receptor. J Clin Endocrinol Metab 83:4481–4489
159. **Oh PS, Kim IS, Moon YH,** A molecular genetic study on the two Korean patients with Laron syndrome. Program of the 81st Annual Meeting of The Endocrine Society, San Diego, CA, 1999, p 140 (Abstract P1-26)
160. **Enberg B, Luthman H, Segnestam K, Ritzén EM, Sundström M, Norstedt G** 2000 Characterisation of novel missense mutations in the GH receptor gene causing severe growth retardation. Eur J Endocrinol 143:71–76
161. **Tauber MT, Porra V, Dastot F, Molinas C, Amselem S, Cholin S, Rochiccioli P, Bieth E** 1998 Heterozygous mutation in the WSXWS equivalent motif of the growth hormone receptor in a child with poor response to growth hormone therapy. Growth Horm IGF Res 8:211–216
162. **Jorge AA, Souza SC, Arnhold IJ, Mendonca BB** 2004 The first homozygous mutation (S226I) in the highly-conserved WSXWS-like motif of the GH receptor causing Laron syndrome: suppression of GH secretion by GnRH analogue therapy not restored by dihydrotestosterone administration. Clin Endocrinol (Oxf) 60:36–40
163. **Kou K, Lajara R, Rotwein P** 1993 Amino acid substitutions in the intracellular part of the growth hormone receptor in a patient with the Laron syndrome. J Clin Endocrinol Metab 76:54–59
164. **Iida K, Takahashi Y, Kaji H, Onodera N, Takahashi MO, Okimura Y, Abe H, Chihara K** 1999 The C422F mutation of the growth hormone receptor gene is not responsible for short stature. J Clin Endocrinol Metab 84:4214–4219
165. **Goddard AD, Dowd P, Chernausek S, Geffner M, Gertner J, Hintz R, Hopwood N, Kaplan S, Plotnick L, Rogol A, Rosenfield R, Saenger P, Mauras N, Hershkopf R, Angulo M, Attie K** 1997 Partial growth-hormone insensitivity: the role of growth-hormone receptor mutations in idiopathic short stature. J Pediatr 131:S51–S55
166. **Saenger PH** 1999 Partial growth hormone insensitivity—idiopathic short stature is not always idiopathic. Acta Paediatr Suppl 88:194–198
167. **Chujo S, Kaji H, Takahashi Y, Okimura Y, Abe H, Chihara K** 1996 No correlation of growth hormone receptor gene mutation P561T with body height. Eur J Endocrinol 134:560–562
168. **Arman A, Ozon A, Isguven PS, Coker A, Peker I, Yordam N** 2008 Novel splice site mutation in the growth hormone receptor gene in Turkish patients with Laron-type dwarfism. J Pediatr Endocrinol Metab 21:47–58
169. **Otsuka T, Iwatani N, Kodama M, Sakakida M, Shichiri M, Jinno Y, Niikawa N, Miike T** 1997 The growth hormone receptor gene mutation of a Japanese patient with Laron syndrome. Jpn J Hum Genet 42:323–329
170. **Berg MA, Guevara-Aguirre J, Rosenbloom AL, Rosenfeld RG, Francke U** 1992 Mutation creating a new splice site in the growth hormone receptor genes of 37 Ecuadorean patients with Laron syndrome. Hum Mutat 1:24–32
171. **Baumbach L, Schiavi A, Bartlett R, Perera E, Day J, Brown MR, Stein S, Eidson M, Parks JS, Cleveland W** 1997 Clinical, biochemical, and molecular investigations of a genetic isolate of growth hormone insensitivity (Laron's syndrome). J Clin Endocrinol Metab 82:444–451
172. **David A, Miraki-Moud F, Shaw NJ, Savage MO, Clark AJ, Metherell LA** 2010 Identification and characterisation of a novel GHR defect disrupting the polypyrimidine tract and resulting in GH insensitivity. Eur J Endocrinol 162:37–42
173. **Shevah O, Nunez O, Rubinstein M, Laron Z** 2002 Intronic mutation in the growth hormone receptor gene in a Peruvian girl with Laron syndrome. J Pediatr Endocrinol Metab 15:1039–1040
174. **Woods KA, Fraser NC, Postel-Vinay MC, Savage MO, Clark AJ** 1996 A homozygous splice site mutation affecting the intracellular domain of the growth hormone (GH) receptor resulting in Laron syndrome with elevated GH-binding protein. J Clin Endocrinol Metab 81:1686–1690
175. **Besson A, Salemi S, Eblé A, Joncourt F, Gallati S, Jorge AA, Mullis PE** 2004 Primary GH insensitivity (Laron syndrome) caused by a novel 4 kb deletion encompassing exon 5 of the GH receptor gene: effect of intermittent long-term treatment with recombinant human IGF-I. Eur J Endocrinol 150:635–642
176. **Meacham LR, Brown MR, Murphy TL, Keret R, Silbergeld A, Laron Z, Parks JS** 1993 Characterization of a noncontiguous gene deletion of the growth hormone receptor in Laron's syndrome. J Clin Endocrinol Metab 77:1379–1383
177. **Gastier JM, Berg MA, Vesterhus P, Reiter EO, Francke U** 2000 Diverse deletions in the growth hormone receptor

gene cause growth hormone insensitivity syndrome. Hum Mutat 16:323–333

178. **Yamamoto H, Kouhara H, Iida K, Chihara K, Kasayama S** 2008 A novel growth hormone receptor gene deletion mutation in a patient with primary growth hormone insensitivity syndrome (Laron syndrome). Growth Horm IGF Res 18:136–142
179. **Gennero I, Edouard T, Rashad M, Bieth E, Conte-Aurio F, Marin F, Tauber M, Salles JP, El Kholy M** 2007 Identification of a novel mutation in the human growth hormone receptor gene (GHR) in a patient with Laron syndrome. J Pediatr Endocrinol Metab 20:825–831
180. **Hwa V, Haeusler G, Pratt KL, Little BM, Frisch H, Koller D, Rosenfeld RG** 2006 Total absence of functional acid labile subunit, resulting in severe insulin-like growth factor deficiency and moderate growth failure. J Clin Endocrinol Metab 91:1826–1831
181. **van Duyvenvoorde HA, Kempers MJ, Twickler TB, van Doorn J, Gerver WJ, Noordam C, Losekoot M, Karperien M, Wit JM, Hermus AR** 2008 Homozygous and heterozygous expression of a novel mutation of the acid-labile subunit. Eur J Endocrinol 159:113–120
182. **Fofanova-Gambetti OV, Hwa V, Kirsch S, Pihoker C, Chiu HK, Högler W, Cohen LE, Jacobsen C, Derr MA, Rosenfeld RG** 2009 Three novel IGFALS gene mutations resulting in total ALS and severe circulating IGF-I/IGFBP-3 deficiency in children of different ethnic origins. Horm Res 71:100–110
183. **Bang P, Fureman A-L, Nilsson A-L, Boström J, Kriström B, Ekström K, Hwa V, Rosenfeld RG, Carlsson-Skwirut C** 2009 A novel missense mutation of the *ALSIGF* gene causing a L172F substitution in LRR6 is associated with short stature in two Swedish children homozygous or compound heterozygous for the mutation. Horm Res 72(Suppl 3): Abstract P01-063:86
184. **Gallego-Gomez E, Sanchez del Pozo J, Cruz Rojo J, Zurita-Muñoz O, Gracia-Bouthelier R, Heath KE, Campos-Barros A** 2009 Novel compound heterozygous *IGFALS* mutation associated with impaired postnatal growth and low circulating IGF-I and IGFBP-3 levels. Horm Res 72(Suppl 3):Abstract P01-076:90–91