Short and tall stature: a new paradigm emerges

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Abstract | In the past, the growth hormone (GH)–insulin-like growth factor 1 (IGF-1) axis was often considered to be the main system that regulated childhood growth and, therefore, determined short stature and tall stature. However, findings have now revealed that the GH–IGF-1 axis is just one of many regulatory systems that control chondrogenesis in the growth plate, which is the biological process that drives height gain. Consequently, normal growth in children depends not only on GH and IGF-1 but also on multiple hormones, paracrine factors, extracellular matrix molecules and intracellular proteins that regulate the activity of growth plate chondrocytes. Mutations in the genes that encode many of these local proteins cause short stature or tall stature. Similarly, genome-wide association studies have revealed that the normal variation in height seems to be largely due to genes outside the GH–IGF-1 axis that affect growth at the growth plate through a wide variety of mechanisms. These findings point to a new conceptual framework for understanding short and tall stature that is centred not on two particular hormones but rather on the growth plate, which is the structure responsible for height gain.

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Introduction

During the past 25 years, a prevalent conceptual framework for understanding short stature and tall stature has been centred on the growth hormone (GH)–insulin-like growth factor 1 (IGF-1) axis.[1–4] Children were thought to grow taller primarily because the pituitary gland produces GH, which stimulates the liver to produce IGF-1. This mindset was understandable historically because the role of the GH–IGF-1 axis in longitudinal bone growth was discovered in the 1950s.[5] In addition, the measurement of these two circulating factors, GH and IGF-1, was readily accomplished by radioimmunoassay in the 1960s and 1970s, respectively.[6,7] Furthermore, GH was historically the main therapeutic approach available to treat patients with short stature.

On the basis of the historical paradigm, short stature has sometimes been divided into defects within the GH–IGF-1 system versus those outside this axis.[8] This thinking also tended to dominate our speculations about the causes of idiopathic short stature. For example, idiopathic short stature was thought to be due to: secondary IGF-1 deficiency (due to subtle disorders of GH secretion); primary IGF-1 deficiency (low serum levels of IGF-1 with normal GH secretion); IGF-1 resistance; or 'other causes'.[1,9] Similarly, the normal variation in height in the general population might be due to subtle modulation of the GH–IGF-1 axis.[10,11]

However, unambiguous defects in the GH–IGF-1 axis can only be identified in a minority of children who present to the clinician with short stature.[12] In some of these children, the underlying molecular defects have been found, including mutations that cause GH deficiency (mutations in *GH1* and *GHRHR*), GH insensitivity or primary IGF-1 deficiency (mutations in *GHR*, *IGFI*, *IGFALS* and *STAT5B*) and IGF insensitivity (mutations in *IGF1R*).[13] However, these genetic abnormalities are rare. Far more children fail GH stimulation tests than have an identifiable mutation; however, this failure seems to result primarily from poor test specificity.[14,15] Similarly, many children with short stature have IGF-1 levels in the lower part of the normal range or even below the normal range.[16] This condition has been labelled primary IGF-1 deficiency. However, it is unclear how many of these children have low IGF-1 levels secondary to poor nutritional intake[17] (for example, due to diminished appetite,[18] subtle chronic disease or[19] other primary disorders[20]) or if these low levels of IGF-1 are simply due to a delay in the physiological increase in IGF-1 that occurs with ageing and puberty, as many children with short stature have delayed maturation of other

Competing interests
J.B. is listed as a co-inventor on a patent application by the NIH for targeted treatment of cartilage disorders. L.S. has received speakers' honoraria and/or research support from Ferring, Merck Serono, Novo Nordisk and Pfizer, and has submitted a patent application for novel peptides to treat bone or cartilage disorders and other diseases. A.D. has been a faculty speaker at continuing medical education symposia sponsored by Ipsen, Novo Nordisk and Sandoz. M.P. has received research support from Novo Nordisk, Pfizer and Teva, personal fees from Novo Nordisk and is a director of NG Solutions. J.M.W. has served as a consultant for Ammonett, Biopartners, Merck Serono, OPKO, Pfizer, Teva and Versartis, and has received speakers' honoraria from Lilly, Merck Serono, Pfizer, Sandoz and Versartis. O.N. has received a European Society for Paediatric Endocrinology research fellowship sponsored by Novo Nordisk and speaker's honoraria from Lilly. F.D.L. has no competing interests to declare.

Key points

- Linear growth (that is, gain in height) is determined by the rate of growth plate chondrogenesis
- Short stature is caused by decreased chondrogenesis whereas tall stature is the result of increased chondrogenesis
- The rate of growth plate chondrogenesis is regulated by many systems, including those related to intracellular, paracrine and extracellular matrix factors, as well as endocrine mechanisms
- Findings from the past decade have identified many new genetic defects responsible for short and tall stature that occur across the systems that regulate growth plate activity
- Similarly, genome-wide association studies have revealed that the normal variation in height seems to be due to many genes that affect the growth plate through a variety of mechanisms
- These new findings suggest a novel conceptual framework for understanding short and tall stature, which is centred on the growth plate—the structure responsible for height gain

physiological processes.[17] Thus, the vast majority of children with short stature do not have a well-substantiated defect in the GH–IGF-1 axis.[12]

In this Review, we explore the latest discoveries in the molecular and cell biology of childhood growth and in the clinical genetics of childhood growth disorders. Taken together, these new findings reveal that the GH–IGF-1 axis is just one of many regulatory systems that control linear growth (gain in height). On the basis of these new findings, we propound a broader conceptual framework to understand linear growth disorders. This more general paradigm is centred on growth plate chondrogenesis, which is the fundamental biological process that drives linear growth in children.

A new paradigm

GH and IGF-1 stimulate linear growth in children by acting on the growth plate (Figure 1). The growth plate is a thin layer of cartilage that is found in most bones (other than the skull and facial bones), including the long bones and vertebrae. In the growth plate, chondrocytes proliferate, undergo hypertrophy and secrete cartilage extracellular matrix components (Figure 1). These processes generate new cartilage tissue, which is subsequently remodelled into bone tissue.[21,22] The net result is that new bone is progressively created at the growth plate, which causes bones to grow longer and children to grow taller. GH acts on the growth plate to stimulate new bone formation both through regulating circulating levels of IGF-1 and also locally, in part through local IGF-1 production.[5,22]

However, findings from both basic and clinical studies (discussed later in this Review) have revealed that the GH–IGF-1 axis is just one of many regulatory systems that control chondrogenesis in the growth plate and, therefore, regulates linear growth in children (Figure 2). Consequently, normal growth in children requires not just concentrations of GH and IGF-1 within the normal range, but also normal production and action of multiple other hormones, paracrine factors and extracellular matrix molecules. Normal growth also requires the multiple intracellular processes needed for chondrocyte proliferation, hypertrophy and extracellular matrix production to function as usual. Many new genes have been identified that, when mutated, result in short stature or tall stature, the majority of which do not participate in the GH–IGF-1 system (these findings are discussed later in this Review). Similarly, normal variation in height seems to be largely due to genes outside the GH–IGF-1 axis that modulate growth at the growth plate through a wide variety of mechanisms.[23] These novel findings point to a new conceptual framework for understanding short and tall stature, a framework that is centred not on two particular hormones but the growth plate itself.

Multiple pathways for normal growth

In the past, the study of childhood growth was severely limited by the available experimental methods. Endocrine factors in the circulation could be measured by immunoassays, but few methods were available to study, for example, paracrine factors that act locally in the growth plate without entering the circulation or intracellular molecular pathways that regulate chondrocyte proliferation and differentiation. However, in the past few decades, a wide variety of new experimental approaches have yielded an explosion of information about the function of the growth plate. For example, knockout of many genes not previously known to be important in the growth plate have produced phenotypes in which skeletal growth at the growth plate is affected, thus uncovering unexpected new areas of growth plate physiology.[21] In parallel, new molecular genetic techniques used in clinical research have been used to identify genetic abnormalities that cause short stature, many of which occur in genes involved, not in the GH–IGF-1 axis, but in other, often local, pathways necessary for a normally functioning growth plate. Taken together, the basic biology and clinical genetic studies have expanded our view of childhood growth physiology and pathophysiology.

Hormones and cytokines

In addition to GH and IGF-1, multiple other hormones also regulate linear growth, including thyroid hormone, glucocorticoids, estrogens and androgens. Evidence suggests that each of these endocrine factors regulate growth in part by a direct action on the growth plate. For example, infusion of dexamethasone, a synthetic glucocorticoid, directly into the growth plate causes local slowing of growth in that growth plate.[24,25] Furthermore, addition of dexamethasone to culture medium slows growth of cultured fetal metatarsal bones.[24,25] Clinically, treatment of glucocorticoid-induced short stature with GH can partially compensate for administering a low dose of glucocorticoid; however, GH treatment has little effect when high doses of glucocorticoids are administered.[26] Similar evidence has been found for direct effects of thyroid hormone,[27,28] androgen[29,30] and estrogen.[31,32] In addition, these hormonal systems can interact. For example, glucocorticoids have complex effects on GH production[33] and inhibit production of thyroid hormone.[34]

Estrogen has complex effects on the growth plate, not only altering growth rate, but also accelerating the

loss of progenitor cells in the resting zone and thereby speeding up the developmental program of growth plate senescence, which causes early cessation of growth.[35,36] Consequently, inactivating mutations in the genes that encode the estrogen receptor[37,38] and aromatase[39] (the enzyme that converts androgens to estrogens) cause the program of growth plate senescence to progress more slowly than in individuals without these mutations, which prolongs linear growth beyond adolescence and thus results in adult tall stature. Aromatase inhibitors produce similar effects and are, therefore, under investigation as a treatment for short stature in boys.[40,41] Some of the estrogen that modulates growth physiologically might be produced locally by chondrocytes in the growth plate that express aromatase, as well as other enzymes that metabolize steroids.[32,42]

Proinflammatory cytokines are endogenously produced by chondrocytes in the growth plate and might act intrinsically to modulate longitudinal bone growth.[43] Furthermore, the growth plate is targeted by extrinsic factors, including cortisol and proinflammatory cytokines, levels of which are both raised during stress and chronic inflammation.[44] Some of these proinflammatory cytokines negatively regulate growth plate function (whether circulating or endogenous proinflammatory cytokines are more physiologically important is currently unclear).[45] Some evidence suggests that tumour necrosis factor, IL-1β and IL-6 act directly on growth plate cartilage to suppress bone growth.[43,46,47] These cytokines might act in synergy, which would further potentiate their negative effects on the function of cartilage in the growth plate.[47]

Longitudinal bone growth is also regulated by nutritional intake, which is mediated in part through a complex endocrine network that includes leptin, IGF-1, sex steroids, thyroid hormone and glucocorticoids.[22] As a result, undernourished children have impaired linear growth (despite raised GH levels) whereas children with obesity have normal or mildly accelerated growth (despite low GH levels).[48]

Physical mechanisms

Growth plate function can also be affected by physical mechanisms. Even fairly low doses of ionizing radiation, such as a single dose of 10 Gy, can impair longitudinal growth.[49] Mechanical compression across the growth plate also impairs the elongation of bones,[50] which is partly due to decreased enlargement of hypertrophic chondrocytes.[51] The inhibitory effects of compression on the growth plate might contribute to progression of scoliosis and tibia vara (also called Blount disease).[50] Similarly, physical compression is used in the clinic to correct inequalities in limb length and angular deformities.[52] However, in children, the effects of dynamic load variation due to exercise has not been well established.[53]

Paracrine factors

One area in which our understanding has advanced enormously involves the role of paracrine signals in the growth plate. Paracrine factors are secreted by chondrocytes in the growth plate, or sometimes cells in the surrounding perichondrium, and act locally on chondrocytes to regulate proliferation and differentiation. The use of *in vitro* and *in vivo* approaches has uncovered a host of paracrine factors necessary for normal growth plate function, including multiple fibroblast growth factors (FGFs),[54–57] bone morphogenetic proteins (BMPs),[58–60] Wnts,[61,62] the parathyroid hormone-related protein (PTHrP) and Indian hedgehog (IHH) pathway[21] and the C-type natriuretic peptide (CNP)–atrial natriuretic peptide receptor 2 (NPR2) pathway.[63] Mutations in genes involved in these paracrine signalling systems can severely impair bone growth in both mice and humans.

For example, fibroblast growth factor receptor 3 (FGFR-3) is a negative regulator of growth plate chondrogenesis.[21] Consequently, activating mutations in *FGFR3* impair elongation of bone in patients with achondroplasia, hypochondroplasia or thanatophoric dysplasia.[64] A report published in 2015 has identified a mutation that activates *FGFR3* in a family with autosomal dominant proportionate short stature.[65] Conversely, heterozygous[66] and homozygous[67] mutations that inactivate *FGFR3* have been reported in individuals with tall stature. At the growth plate, FGF signalling via FGFR-3 leads to activation of several pathways, including the mitogen activated protein kinase (MAPK) and Janus kinase–signal transducer and activator of transcription (JAK–STAT) pathways.[68,69] FGFR-3 signalling negatively regulates growth by decreasing proliferation

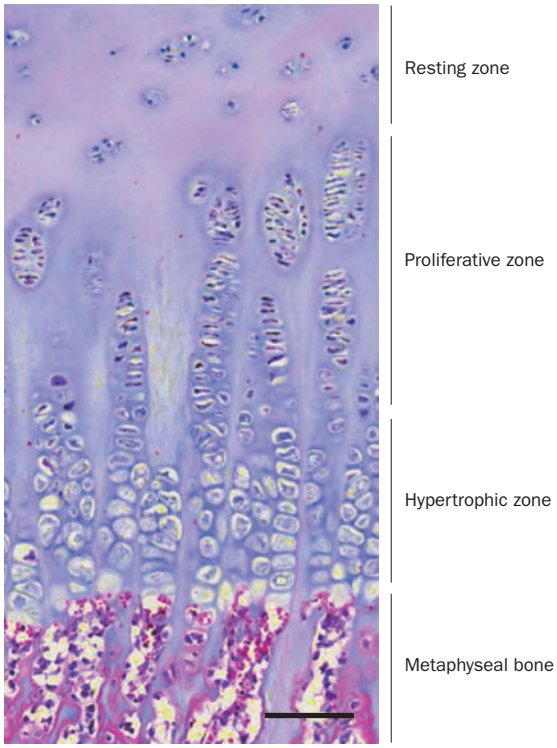

Figure 1 | Human growth plate histology from an 11-year old boy. The growth plate comprises three histologically and functionally distinct zones—the resting, proliferative and hypertrophic zones—sandwiched between the epiphyseal bone and the metaphyseal bone. Bar represents 100 μm.

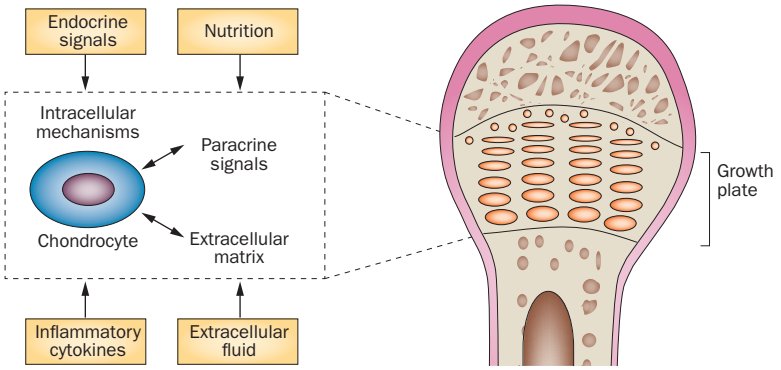

Figure 2 | Regulation of growth plate function. Chondrocyte proliferation and differentiation in the growth plate are regulated by many factors, including nutritional, endocrine, inflammatory cytokines, extracellular fluid (for example, oxygen and pH), paracrine, extracellular matrix and intracellular mechanisms. Not depicted are the interactions among many of these systems; for example, nutritional intake strongly affects endocrine regulators of the growth plate.

in the proliferative zone, decreasing production of the extracellular matrix, accelerating the onset of hypertrophic differentiation and decreasing the size of the hypertrophic chondrocytes.[57,70–72] These effects are at least in part due to interactions with other paracrine factors, including downregulation of IHH expression, as well as interactions with CNP and BMP signalling.[72–74]

The PTHrP and IHH paracrine system also has a critical role in the regulation of growth plate function. These two paracrine factors form a negative feedback loop within the growth plate that regulates chondrocyte hypertrophy and proliferation.[21] Consequently, mutations in the genes that encode IHH, PTHrP and parathyroid hormone receptor 1 (PTH1R) cause specific skeletal dysplasias. For example, heterozygous mutations in *PTHLH*, the gene that encodes PTHrP, result in short stature and short fingers in individuals with brachydactyly, type E2.[75]

Another paracrine factor of importance in the growth plate is CNP. This peptide was named owing to its structural similarity to atrial natriuretic peptide, but has a very different physiologic role as a local positive regulator of growth plate function.[76–78] Consequently, homozygous inactivating mutations in *NPR2*, the gene that encodes the receptor for CNP, cause a severe skeletal dysplasia, termed Moroteaux acromesomelic dysplasia.[79] Interestingly, heterozygous mutations cause a milder phenotype than homozygous mutations, presenting as short stature without clear signs of a skeletal dysplasia.[20] Studies published over the past 2 years suggest that ~2% of children who present with idiopathic short stature have mutations in *NPR2*.[80–82] Conversely, overexpression of CNP[83,84] or activating mutations in *NPR2*[85,86] result in tall stature. Binding of CNP to NPR2 stimulates the guanylyl cyclase activity of the receptor, thereby increasing synthesis of cGMP, which activates the type II cGMP-dependent protein kinase.[87] Mice deficient in this protein kinase also have severely impaired growth plate function.[88] This signalling system leads to inhibition of the MAPK pathway, thus antagonizing FGFR signalling,[89] which provides an explanation for the beneficial effects of CNP overexpression or administration in a mouse model of achondroplasia.[90]

BMPs, also known as growth and differentiation factors (GDFs), belong to the transforming growth factor β (TGF-β) superfamily of paracrine factors. When they were originally discovered, BMPs were described as the component of demineralized bone matrix that can induce ectopic bone formation; however, they were subsequently found to also regulate a multitude of processes in skeletal development, including spatial regulation of proliferation and differentiation in the growth plate.[63] Consistent with this finding, inactivating mutations in the genes that encode several BMPs, their receptors and antagonists lead to skeletal dysplasias, including brachydactyly, type A2 (*BMP2* or *BMPR1B*), brachydactyly, type A1 and brachydactyly, type C (*GDF5*), chondrodysplasia, Grebe type (*GDF5*), Klippel–Feil syndrome 1 (*GDF6*) and proximal symphalangism 1A (*NOG*). Local modulation by antagonists and sequestration by extracellular matrix molecules are crucial for local regulation of BMP and TGF-β signalling during development and growth. Consequently, excessive TGF-β signalling has been identified as an important pathogenic mechanism for Marfan syndrome,[91] which is characterized by disproportionate skeletal overgrowth and aortic dilation[92] and is caused by mutations in the gene that encodes fibrillin 1 (*FBN1*). Angiotensin II receptor blockers act as TGF-β antagonists and are, therefore, being evaluated in clinical studies for prevention of aortic dilatation in Marfan syndrome.[93]

Cartilage extracellular matrix

Chondrocytes secrete a unique extracellular matrix that contains specific collagens (including collagen type II and X), proteoglycans (including aggrecan) and noncollagenous proteins that are vital for normal functioning of the growth plate. This extracellular matrix provides the compressible, resilient structural properties of cartilage and also interacts with signalling molecules to regulate growth plate chondrogenesis.[94] As a result, mutations in genes that encode matrix proteins and proteoglycans often interfere with growth plate function.[95] For example, mutations in *COL10A1*, the gene that encodes collagen α-1(X) chain, cause a skeletal dysplasia termed metaphyseal chondrodysplasia, Schmid type.[96]

Mutations in the gene *ACAN*, which encodes aggrecan (a major proteoglycan component of the cartilage extracellular matrix), also affect linear growth. Homozygous mutations lead to a severe skeletal dysplasia, spondyloepimetaphyseal dysplasia aggrecan type.[97] Heterozygous mutations can present as a milder skeletal dysplasia, (spondyloepiphyseal dysplasia, Kimberley type[98]) or as short stature without radiographic evidence of skeletal dysplasia, which can either be disproportionate[99] or proportionate.[100] The short stature is typically associated with an advanced bone age and early cessation of growth.[100] In some patients, this disorder affects not only the growth plate cartilage but also the articular cartilage, which causes osteochondritis dissecans and early-onset osteoarthritis.[99,100] In animal models, mutations in *Acan* cause

growth plate dysfunction, including decreased chondrocyte proliferation and accelerated hypertrophic chondrocyte differentiation associated with disrupted IHH, FGF and BMP signalling.[101,102]

Other noncollagenous matrix proteins might also interact with paracrine signals. For example, biglycan and decorin modulate growth and bone formation through interactions with TGF-β, as demonstrated by the postnatal growth retardation and osteoporosis seen in mice lacking biglycan.[103] Similarly, excessive TGF-β signalling has been identified as an important pathogenic mechanism in Marfan syndrome.[91,92] Interestingly, mutations in *FBN1* also cause short stature in Weill–Marchesani syndrome, geleophysic dysplasia and acromicric dysplasia.[104]

Intracellular pathways

A variety of intracellular pathways that have important roles in growth plate chondrogenesis have also been discovered. For example, transcription factors SOX-5, SOX-6 and SOX-9 are critical regulators of chondrocyte differentiation.[105] As one would expect, mutations that affect key intracellular pathways lead to growth disorders in humans. For example, inactivating mutations in *SOX9* result in a severe skeletal dysplasia, campomelic dysplasia.[105]

As another example, homozygous inactivating mutations in *SHOX*, which encodes another transcription factor expressed in the growth plate,[106] are associated with Langer mesomelic dysplasia (characterized by severe defects in bone growth). Heterozygous inactivating mutations, or deletions of *SHOX* or its enhancer regions, result in a milder skeletal dysplasia (Leri–Weill dyschondrosteosis) or can present clinically as idiopathic short stature, with body proportions that are mildly affected or sometimes within the normal range.[107] Of individuals presenting with idiopathic short stature, 2–15% have mutations in *SHOX*, with the exact percentage depending on the study.[108] Conversely, increased copies of *SHOX* are associated with tall stature in individuals with Klinefelter syndrome and other types of sex chromosome aneuploidy.[109]

Another pathway that is important for skeletal growth due to its effect on cellular proliferation and differentiation of growth plate chondrocytes is the Ras–MAPK signalling pathway. This pathway integrates signals from several growth factors including FGFs, CNP and epidermal growth factor.[68] Ras, a small GTPase, signals via MAPK cascades to phosphorylate numerous cytoplasmic and nuclear proteins, regulating cell proliferation and differentiation.[110] Increased activation of this pathway results in a number of overlapping syndromes including Noonan syndrome, LEOPARD syndrome, Costello syndrome, cardiofaciocutaneous syndrome and neurofibromatosis–Noonan syndrome, which are all characterized by neurocutaneous manifestations and varying degrees of postnatal growth failure.[111,112] By contrast, Sotos syndrome (characterized by tall stature) is associated with decreased activity of the Ras–MAPK pathway.[113]

Skeletal growth is also regulated by nuclear factor κB (NF-κB) and a group of seven transcription factors, including transcription factor p65 (RelA), c-Rel, RelB, p50/p105 (NF-κB1) and p52/p100 (NF-κB2). In growth plate chondrocytes, NF-κB–p65 helps to mediate the stimulatory effects of GH and IGF-1 on chondrogenesis.[114] In humans, heterozygous loss-of-function mutations in the gene that encodes IκBα, an essential component of the NF-κB pathway, result in GH insensitivity and growth failure, as well as ectodermal dysplasia and immunodeficiency.[115]

Mutations in genes that encode proteins involved in fundamental cellular processes can produce severe global growth deficiencies, termed primordial dwarfisms, that affect not just the growth plate but also multiple other tissues and typically impair both prenatal and postnatal growth.[116] For example, 3M syndrome, which includes severe intrauterine growth retardation and postnatal short stature, is caused by defects in one of three genes: *CUL7*,[117] *OBSL1*[118] and *CCDC8*.[119] The products of these three genes form a complex that has a critical role in maintaining microtubule integrity; defects in these genes lead to aberrant cell division.[120] Similarly, mutations in a centrosomal protein (pericentrin) are associated with microcephalic osteodysplastic primordial dwarfism type II.[121] Other centrosomal proteins (such as CENP-J, CEP152 and CEP63) are implicated in Seckel syndrome.[116] Mutations in the DNA origin recognition complex underlie Meier–Gorlin syndrome,[122,123] and defects in DNA damage repair underlie growth disorders such as Cockayne syndrome and Bloom syndrome.[124]

Interestingly, tall stature can be caused by mutations in genes that control epigenetic modifications, including DNA and histone methylation, and thereby chromatin formation and gene expression. For example, heterozygous mutations in DNA methyltransferase 3A (*DNMT3A*) cause tall stature, a distinctive facial appearance and intellectual disability.[125] Similarly, heterozygous mutations in *EZH2*, which encodes an enzyme that specifically methylates lysine residue 27 of histone 3 (H3K27, which is associated with transcriptional repression), are associated with Weaver syndrome (characterized by prenatal and postnatal overgrowth and a markedly advanced bone age).[126] In addition, most cases of Sotos syndrome result from mutations in *NSD1*; the protein product of this gene is a methyltransferase that methylates histone H3 lysine 36 and other substrates and also interacts with nuclear hormone receptors, thereby regulating transcription of target genes.[113]

CNV and short stature

Findings published in the past 3 years suggest that ~10% of patients with idiopathic short stature carry a disease-causing copy number variation (CNV).[127–129] The phenotype of patients with CNVs includes: growth failure of both prenatal onset and postnatal onset; both proportionate and disproportionate short stature; and both syndromic and nonsyndromic short stature.[128,129] However, in individual patients, knowing whether the growth failure was due to the CNV is difficult; therefore, additional studies are needed to improve definitions of the prevalence and phenotypes of CNV-associated short stature.

Box 1 | Aetiology of short stature

Intrinsic to growth plate

Paracrine
- C-type natriuretic peptide signalling (deficiency)
- Fibroblast growth factor signalling (excess)
- Parathyroid hormone-related protein signalling (deficiency, excess)

Cartilage extracellular matrix
- Abnormal structural properties
- Effects on growth factor signalling

Intracellular
- Chondrocyte transcription factors (deficiency)
- Ras–MAPK signalling (deficiency)

Extrinsic to growth plate

Nutritional intake
- Calories (deficiency)
- Protein (deficiency)
- Zinc (deficiency)
- Vitamin A (excess)

Endocrine
- Growth hormone (deficiency or insensitivity)
- Insulin-like growth factor 1 (deficiency or insensitivity)
- Thyroid hormone (deficiency or insensitivity)
- Glucocorticoid (excess)
- Estrogen (excess results in early growth termination)
- Androgen (deficiency)

Inflammatory cytokines
- IL-1β (excess)
- IL-6 (excess)
- Tumour necrosis factor (excess)

Extracellular fluid
- Oxygen (deficiency)
- Acidosis

Physical factors
- Mechanical trauma
- Ionizing radiation

This list provides examples of disorders affecting linear growth, but is not exhaustive.

Normal variation in adult height

The normal variation in human stature is well known to have a large genetic component and a polygenic pattern of inheritance. The specific genes involved have only begun to be elucidated in the past few years (primarily since 2008). A large meta-analysis of genome-wide association studies identified 423 loci that contribute to the normal variation in adult stature.[130] Presumably, each locus contains at least one gene in which common polymorphisms affect linear growth in humans. Although the precise gene within each locus that is responsible for the effect typically cannot be pinpointed with certainty, evidence indicates that a large number of the causative genes are expressed in, and function in, the growth plate.[130,131] These genes include multiple genes involved in paracrine signalling by the PTHrP–IHH feedback loop, BMPs, FGFs, Wnts/β-catenin and CNP. These loci also contain a much smaller number of genes known to participate in the GH–IGF-1 axis, including *GH1* (encodes GH), *GHSR* (encodes the GH secretagogue receptor) and *IGF1R* (encodes the principal receptor for IGF-1).[130,131] In our opinion, many children with short stature, particularly those with mild short stature and with a pedigree that suggests a polygenic inheritance, are likely to have inherited multiple polymorphisms that negatively modulate linear growth associated with short stature. Interestingly, a study published in 2014 that included a large cohort of extremely tall individuals of European descent reported that common sequence variants are also associated with tall stature.[132]

A broader conceptual framework

The findings discussed in this Review necessitate a new framework with which to conceptualize short stature. The categorization of idiopathic short stature into subtypes of GH deficiency, GH insensitivity, IGF-1 deficiency and IGF-1 insensitivity, which seemed reasonable when we could only measure hormones and knew little about the causes of short stature, is now not nearly broad enough. Using this system would require us to jam all the myriad abnormalities involving intracellular, extracellular matrix and paracrine signalling defects into the category 'IGF-1 insensitivity'. Instead, the large picture emerging from the latest findings acknowledges that GH and IGF-1 are important factors for growth, but also that they are only two of many factors that influence growth plate function and, therefore, human growth.

A broader conceptual framework can now be formulated that is centred, not on the GH–IGF-1 axis, but on the growth plate (Figure 2). Short stature is caused by growth plate dysfunction that can result either from a primary defect (that is, a disorder intrinsic to the growth plate) or a secondary defect, in which the growth plate is adversely affected by a disorder elsewhere in the body (Box 1, Figure 2). Primary defects might involve: paracrine signalling systems in the growth plate; cartilage extracellular matrix molecules; or intracellular pathways in growth plate chondrocytes. In secondary disorders, chondrocytes in the growth plate can be adversely affected through a variety of mechanisms, including nutritional effects (mediated in part by endocrine signals), endocrine signalling, inflammatory cytokines, extracellular fluid (such as acidosis) and physical factors (such as radiation).

These observations provide a conceptual framework that is based on the underlying biological mechanisms. For some disorders, including many dysmorphic syndromes, constitutional delay of growth and idiopathic short stature, the mechanism responsible for growth plate dysfunction remains unknown. A classification scheme proposed by the European Society for Paediatric Endocrinology, which also divides the causes of short stature into primary and secondary defects, is less mechanism-oriented than the framework based on the growth plate but is useful for practical clinical purposes.[133] Although growth disorders can be environmental or polygenic in aetiology, many growth disorders arise from single gene defects (Tables 1 and 2).

A genotype–phenotype spectrum

Previously, skeletal dysplasias and idiopathic short stature were considered to be largely distinct entities. However, the latest findings indicate that defects in the same genes, such as *SHOX*, *NPR2*, *ACAN* and *FGFR3*, can present clinically either as a skeletal dysplasia or as idiopathic

Table 1 | Single gene defects intrinsic to growth plate that cause disorders of childhood growth

Mechanism	Gene	Effect on protein	Disorder	Effect on linear growth	Inheritance
Paracrine					
C-type natriuretic peptide signalling	*NPR2*	Loss-of-function	ISS	Short stature	AD*
			Moroteaux acromesomelic dysplasia	Short stature	AR*
		Gain-of-function	Overgrowth with or without skeletal deformities	Tall stature	AD*
Fibroblast growth factor signalling	*FGFR3*	Gain-of-function	Hypochondroplasia, achondroplasia, thanatophoric dysplasia, ISS	Short stature	AD
		Loss-of-function	CATSHL syndrome	Tall stature	AD
Parathyroid hormone-related protein signalling	*GNAS* *PTH1R*	Loss-of-function	Albright hereditary osteodystrophy	Short stature	AD
			Blomstrand chondrodysplasia	Short stature	AR
		Gain-of-function	Jansen metaphyseal dysplasia	Short stature	AD
Cartilage extracellular matrix					
Extracellular matrix structure or function	*ACAN*	Abnormal structure	ISS	Short stature	AD*
			Spondyloepiphyseal dysplasia type Kimberley	Short stature	AD*
			Spondyloepimetaphyseal dysplasia aggrecan type	Extreme short stature	AR*
Extracellular matrix structure or function	*COL2A1*	Abnormal structure	Multiple skeletal dysplasias	Short stature	AD
Extracellular matrix structure or function	*COL10A1*	Abnormal structure	Metaphyseal chondrodysplasia, Schmid type	Short stature	AD
Extracellular matrix structure or function	*COMP*	Abnormal structure	Pseudoachondroplasia	Short stature	AD
Extracellular matrix structure or function	*FBN1*	Abnormal structure	Marfan syndrome	Tall stature	AD
Intracellular					
Transcription factors	*SOX9*	Loss-of-function	Campomelic dysplasia	Short stature	AD
Transcription factors	*SHOX*	Loss-of-function	ISS	Short stature	AD*
			Leri–Weill dyschondrosteosis	Short stature	AD*
			Langer mesomelic dysplasia (homozygous)	Extreme short stature	AR*
Microtubule function	*CUL7*	Loss-of-function	3M syndrome 1	Short stature	AR
Microtubule function	*OBSL1*	Loss-of-function	3M syndrome 2	Short stature	AR
Ras–MAPK signalling	*PTPN11*, *KRAS*, others	Gain-of-function	Noonan syndrome	Short stature	AD
Epigenetic defects	*NSD1*	Loss-of-function	Sotos syndrome	Tall stature	AD
Epigenetic defects	*EZH2*	Uncertain	Weaver syndrome	Tall stature	AD
Epigenetic defects	*DNMT3A*	Loss-of-function	DNMT3A overgrowth syndrome	Tall stature	AD

This list provides examples of single gene defects affecting linear growth, but is not exhaustive. *Mutations in these genes can be considered semidominant in that heterozygous mutations produce a mild phenotype while homozygous mutations result in a more severe phenotype. Abbreviations: AD, autosomal dominant; AR, autosomal recessive; CATSHL, camptodactyly, tall stature and hearing loss; ISS, idiopathic short stature.

short stature.[56,81,100,108] The severe phenotype tends to occur when the gene involved is critical for growth plate function, the mutation severely alters protein function and/or the mutation occurs in the homozygous state. By contrast, the milder phenotype (that is, short stature with normal bone morphology) tends to occur when the gene involved is not critical for growth plate function, the mutation only partially disrupts protein function and/or when the mutation occurs in the heterozygous state (Figure 3).[56,81,100,108] Furthermore, the results from genome-wide association studies suggest that stature in the lower part of the normal range can also be considered part of this spectrum as it seems to be partially due to polymorphisms in the same genes in which mutations cause skeletal dysplasia, such as *COL10A1* (mutations are responsible for metaphyseal chondrodysplasia, Schmid type), *ACAN* (spondyloepimetaphyseal dysplasia, aggrecan type) and *HOXD13* (brachydactyly, type E).[131] Thus, polymorphisms that have a mild effect on protein function or expression tend to result in stature in the lower part of

Table 2 | Single genetic variants extrinsic to growth plate that cause childhood growth disorders*

Endocrine mechanism	Gene	Disorder	Effect on linear growth	Inheritance
GH signalling	*GH1*	Isolated GH deficiency	Short stature	AR, AD
	GHRHR	Isolated GH deficiency	Short stature	AR
	POU1F1, *PROP1*, *LHX3*, others	Combined pituitary hormone deficiency	Short stature	AR
	GHR	GH insensitivity syndrome	Short stature	AR
	STAT5B	Growth hormone insensitivity with immune dysfunction	Short stature	AR
IGF-1 signalling	*IGF1*	IGF-1 deficiency	Short stature	AD, AR
	IGF1R	IGF resistance	Short stature	AD, AR
	ALS	ALS deficiency	Short stature	AR
Thyroid hormone signalling	*TSHR*, *PAX8*, *NKX2-1*, *FOXE1*, *NKX2-5*	Thyroid dysgenesis	Short stature	AD, AR
	SLC5A5, *TP*, *DUOX2*, *SLC26A4*, *TG*, *IYD/DEHAL1*	Thyroid dyshormonogenesis	Short stature	AR
	THRB	Thyroid hormone resistance	Short stature	AD, AR
	THRA	Thyroid hormone resistance	Short stature	AD
Glucocorticoid signalling	*PRKAR1A*	Carney complex (including glucocorticoid excess)	Short stature	AD
	MC2R	Familial glucocorticoid deficiency	Tall stature	AR
Estrogen signalling	*ESR1* (estrogen receptor)	Estrogen resistance	Tall stature	AR
	CYP19A1 (aromatase)	Estrogen deficiency	Tall stature	AR

This list provides examples of single gene defects affecting linear growth, but is not exhaustive. *All these genetic variants have loss-of-function effects on the proteins. Abbreviations: AD, autosomal dominant; AR, autosomal recessive; GH, growth hormone; IGF, insulin-like growth factor.

the normal range. Mildly deleterious and/or heterozygous mutations tend to present as idiopathic short stature or a mild skeletal dysplasia. Strongly deleterious and/or homozygous mutations tend to lead to severe skeletal dysplasia (Figure 3).

In some cases, tall stature can involve the same genes as short stature; however, in tall stature, the mutation has the opposite functional effect on the gene product (Figure 3). For example, as discussed earlier in this Review, homozygous inactivating mutations in *NPR2* result in a skeletal dysplasia with severe short stature, heterozygous inactivating mutations present as a milder skeletal dysplasia or idiopathic short stature and activating mutations in *NPR2* cause tall stature.[85,86] Another example mentioned previously is *FGFR3*. Mutations that activate this gene are associated with proportionate short stature, hypochondroplasia, achondroplasia and thanatophoric dysplasia, whereas inactivating mutations have been reported to lead to tall stature.[66,67] Similarly, loss-of-function mutations or decreased copy numbers of *SHOX* cause short stature, whereas an increased copy number (including in Klinefelter syndrome) is associated with tall stature.

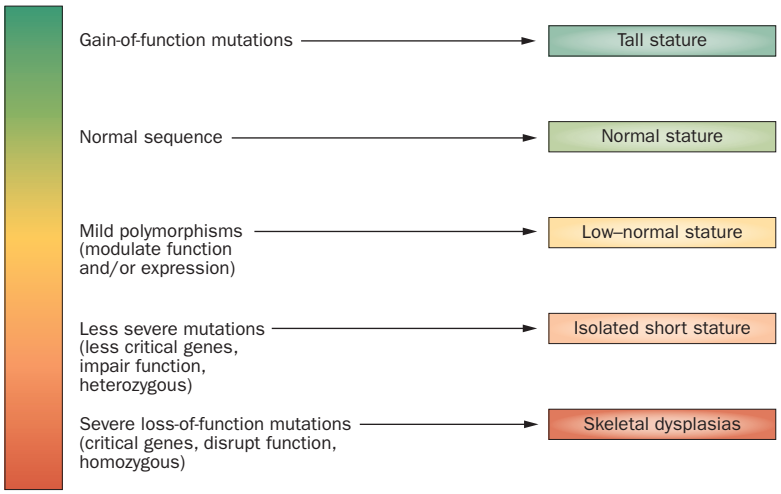

Figure 3 | The phenotypic spectrum caused by genetic variants in genes that regulate growth plate chondrogenesis. This spectrum applies to genes with products that promote longitudinal bone growth, such as *NPR2*. For genes that inhibit longitudinal bone growth, such as *FGFR3*, the spectrum is reversed in that gain-of-function mutations cause short stature while loss-of-function causes tall stature.

Temporal and spatial growth patterns

Some genetic defects, such as activating mutations in *FGFR3* that cause achondroplasia, present with short stature at birth, whereas other genetic defects, such as heterozygous inactivating mutations in *SHOX* that cause Leri–Weill dyschondrosteosis, often result in a normal birth length with short stature developing postnatally. In some cases, this difference might be due to the severity of the growth plate abnormality. For example, severe activating mutations in *FGFR3* associated with thanatophoric dysplasia and achondroplasia cause growth failure of prenatal onset,[134,135] whereas a milder mutation that causes hypochondroplasia often produces a birth length within the normal range with short stature developing postnatally.[136] However, in other cases, the timing of onset (prenatal versus postnatal) might depend on whether the gene is important in the fetal growth plate, which is morphologically and functionally somewhat different from the growth plate in childhood.

As described previously, some defects in genes required for growth plate function produce proportionate short stature whereas others produce disproportionate short stature. Presumably, disproportionate short stature indicates that the gene product involved has a more critical role in some growth plates than in others. Many genetic defects tend to affect growth plates in the long bones more than growth plates in the vertebrae, causing a disproportionate short stature with a greater reduction in leg and arm lengths than in trunk length.[137] We speculate that this common pattern might reflect the fact that the growth plates in the long bones function at a much greater pace than each of the individual growth plates in vertebrae and thus might have increased susceptibility to dysregulation. However, some disorders, such as brachyolmia, tend to affect the spine more than the long bones, causing disproportionate short stature with a short trunk.[138] Brachyolmia is genetically heterogeneous, including mutations in *PAPSS2*, which encodes a sulphotransferase that is required for sulphation of a variety of molecules, such as cartilage glycosaminoglycans and dehydroepiandrosterone.[139]

Mutations in some genes impair development and/or function not only of the growth plate but also of non-skeletal structures, which results in associated congenital anomalies, that is, syndromic short stature. For example, patients with Noonan syndrome, which is caused by mutations in genes involved in the Ras–MAPK pathway such as *PTPN11*, often have short stature associated with distinctive facies, cardiac and renal anomalies, developmental delay and/or coagulation defects.[140] However, mutations in *PTPN11* have also been reported in patients who presented with short stature and were not recognized to have Noonan syndrome prior to sequencing, which suggests that *PTPN11* mutations should be considered in the differential diagnosis of idiopathic short stature.[141]

GH therapy for short stature

Although most short stature results from defects outside the GH–IGF-1 system, treatment with recombinant GH is still somewhat effective in accelerating linear growth in many children with short stature. For example, GH treatment increases the linear growth rate of children with SHOX deficiency,[142] Noonan syndrome[143] and idiopathic short stature.[144,145] Indeed, endogenous GH excess due to a pituitary adenoma can lead to remarkably tall stature in otherwise normal children.[146] These findings indicate that the stimulatory effect of GH on growth plate chondrocyte proliferation and hypertrophy, which is partly mediated by increased levels of IGF-1,[147–149] can, in many cases, non-specifically accelerate linear growth and thereby partially compensate for unrelated molecular defects that affect the growth plate.

Conclusions

During the past 25 years, a prevalent conceptual framework for understanding short and tall stature was centred on the GH–IGF-1 axis. However, findings in basic molecular and cellular biology and in clinical genetics have uncovered a wide range of other regulatory systems that control skeletal growth and an accompanying vast array of genetic defects outside the GH–IGF-1 axis that can lead to disorders of linear growth. As a result, the traditional view of short or tall stature that is centred on the GH–IGF-1 axis is now far too narrow to encompass the ever-growing catalogue of defects that result in abnormal linear growth. A much broader conceptual framework can be based on the simple concept that linear growth disorders necessarily are due to dysfunction of the skeletal growth plate—the structure responsible for bone elongation and, therefore, overall body size. Consequently, short stature can be conceptualized as either a primary or secondary disorder of the growth plate chondrocytes. A related concept that has emerged in the past few years is that sequence variants in genes that affect growth plate function can produce a phenotypic spectrum that ranges from a severe skeletal dysplasia to disproportionate or proportionate short stature, to normal variation in height, to tall stature (Figure 3).

High-throughput sequencing approaches will probably continue to expand the list of genetic defects that are associated with growth plate dysfunction and disorders of linear growth. The clinical application of these broad sequencing approaches will enable physicians to identify the aetiology of growth failure from among the myriad possibilities. We can, therefore, anticipate that the number of children who receive the unhelpful diagnosis of idiopathic short stature will continue to diminish. With these advances, we can look forward to treatment approaches that are tailored to the specific genetic cause of the disorder.

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J.B., F.D.L., A.D., J.M.W. and O.N. researched data for the article, made substantial contributions to discussion of the content, wrote the article and reviewed/edited the manuscript before submission. L.S. made substantial contributions to discussion of the content, wrote the article and reviewed/edited the manuscript before submission. M.P. made substantial contributions to discussion of the content and reviewed/edited the manuscript before submission.