Overgrowth syndromes — clinical and molecular aspects and tumour risk

Frédéric Brioude[1]*, *Annick Toutain*[2], *Eloise Giabicani*[1], *Edouard Cottereau*[3], *Valérie Cormier-Daire*[4] *and Irene Netchine*[1]

Abstract | Overgrowth syndromes are a heterogeneous group of rare disorders characterized by generalized or segmental excessive growth commonly associated with additional features, such as visceromegaly, macrocephaly and a large range of various symptoms. These syndromes are caused by either genetic or epigenetic anomalies affecting factors involved in cell proliferation and/or the regulation of epigenetic markers. Some of these conditions are associated with neurological anomalies, such as cognitive impairment or autism. Overgrowth syndromes are frequently associated with an increased risk of cancer (embryonic tumours during infancy or carcinomas during adulthood), but with a highly variable prevalence. Given this risk, syndrome-specific tumour screening protocols have recently been established for some of these conditions. Certain specific clinical traits make it possible to discriminate between different syndromes and orient molecular explorations to determine which molecular tests to conduct, despite the syndromes having overlapping clinical features. Recent advances in molecular techniques using next-generation sequencing approaches have increased the number of patients with an identified molecular defect (especially patients with segmental overgrowth). This Review discusses the clinical and molecular diagnosis, tumour risk and recommendations for tumour screening for the most prevalent generalized and segmental overgrowth syndromes.

[1]*Sorbonne Université, INSERM UMR_S938, Centre de Recherche Saint Antoine, AP-HP Hôpital Trousseau, Paris, France.*

[2]*CHU de Tours, Hôpital Bretonneau, Service de Génétique, INSERM UMR1253, iBrain, Université de Tours, Faculté de Médecine, Tours, France.*

[3]*CHU de Tours, Hôpital Bretonneau, Service de Génétique, Tours, France.*

[4]*Service de génétique clinique, Université Paris Descartes-Sorbonne Paris Cité, INSERM UMR1163, Institut Imagine, Hôpital Necker-Enfants Malades, Paris, France.*

**e-mail: frederic.brioude@aphp.fr*

https://doi.org/10.1038/s41574-019-0180-z

Overgrowth syndromes are a heterogeneous group of disorders characterized by excessive growth. Overgrowth is usually observed during fetal life (on the basis of ultrasound scans during pregnancy), resulting in excessive length and/or weight at birth (that is, >90th percentile or 2 s.d. above the mean weight and/or length for their gestational age at birth). However, overgrowth can appear later in life. For example, some patients with Beckwith–Wiedemann syndrome (BWS; especially those with imprinting centre 2 (IC2) loss of methylation (LOM) (we describe the molecular mechanism of BWS later in this Review)) can present with tall stature during childhood despite a normal birthweight or length[1].

Historically, all conditions with excessive growth of the whole body were termed overgrowth syndromes (generalized overgrowth). In addition, syndromes associated with segmental overgrowth (one or several parts of the body), such as *PTEN*-related or *PIK3CA*-related syndromes, can be included in this group of disorders as they share common molecular mechanisms or implicate common pathways and expose the affected individual to an increased risk of tumours.

Fetal and postnatal regulation of growth is a complex process involving various factors, including factors with a genetic and/or epigenetic, endocrine and metabolic origin. In addition, trans-placental exchange of nutrients and/or oxygen during pregnancy and fetal exposure to exogenous factors, such as toxins, pollutants or infections, are also linked to fetal growth[2]. When overgrowth results from a metabolic imbalance (for example, in the children of mothers with gestational diabetes mellitus or with pre-existing type 1 or type 2 diabetes mellitus), or as a consequence of a high familial growth potential (constitutional tall stature), overgrowth is usually the only symptom[3]. In cases where epigenetic and/or genetic factors are the cause of overgrowth, additional signs are usually observed, including a large range of dysmorphic features and possibly cognitive impairment or behaviour anomalies. Despite progress during the past 10 years in the identification of epigenetic and genetic aetiologies in patients with overgrowth (especially with the help of next-generation sequencing technologies), up to 50% of patients with syndromic overgrowth still have no identified molecular anomalies[4].

If the molecular factors involved in the control of the cell cycle are disrupted as a somatic event, this can lead to excessive cellular proliferation in a tissue or in a tumour. If the same molecular factors are disrupted by a germinal event, the result can be either overgrowth or growth retardation. For example, the cyclin D kinase

Key points

- Overgrowth syndromes are a heterogeneous group of disorders with clinical overlap and specific clinical traits that make it possible to distinguish between them.
- Most overgrowth syndromes are caused by anomalies in factors that are implicated in the control of cell proliferation or in the control of epigenetic markers.
- Advances in the past decade have enabled the identification of mosaic molecular defects in hyperplastic tissues of patients with segmental overgrowth, particularly in the PI3K–AKT pathway.
- An increased risk of tumours is usually reported in patients with overgrowth syndromes.
- Syndrome-specific tumour screening programmes are needed on the basis of international consensus meetings.
- Strategies for molecular explorations should be based on an accurate clinical description, as the molecular defects can be genetic (mutations), cytogenetic (large rearrangements) or epigenetic.

inhibitor 1C gene (*CDKN1C*; previously called *p57kip2*) is directly involved in the transition from the G1 to the S phase of the cell cycle[5]. *CDKN1C* is underexpressed in various types of tumour (which might explain their excessive proliferation)[6], and germinal mutations of *CDKN1C* lead to BWS in cases of loss-of-function mutations[7] or Silver–Russell syndrome or IMAGe syndrome (two conditions with growth retardation) if a gain-of-function mutation is involved[8,9]. Furthermore, somatic mutations in oncogenes (such as those with protein products involved in the PLAG1–HMGA2–IGF2 pathway), anti-oncogenes or genes of signalling pathways (such as the PI3K–AKT–mTOR pathway) are frequently observed in tumours[10,11]. Germinal mutations in these same oncogenic pathways have now been identified in several conditions characterized by either growth retardation or overgrowth[10,11]. Finally, abnormal epigenetic markers, such as abnormalities in DNA methylation or histone tail modifications, microRNA expression or mRNA processing, are frequently observed in tumours. Intriguingly, germinal mutations in factors that are involved in the regulation of epigenetic markers or RNA processing represent many of the molecular defects identified in syndromic overgrowth[4].

In this Review, we focus on overgrowth syndromes that have a genetic or epigenetic aetiology, their clinical presentation and their molecular mechanisms. Each overgrowth syndrome has a specific set of clinical symptoms that make it possible to differentiate them from each other. As many of these syndromes are associated with an increased risk of tumours, we discuss the screening schedules that have been proposed for each condition. Finally, considering the main molecular mechanisms, we discuss the strategies for molecular investigations in patients with an overgrowth syndrome.

Overgrowth of endocrine origin

Longitudinal growth of bone is stimulated by several hormones, including thyroid hormones, growth hormone (GH), insulin-like growth factor 1 (IGF1) and sex steroids (testosterone and oestradiol)[12]. Therefore, excessive secretion of these hormones (resulting in hyperthyroidism for thyroid hormones, acromegalo-gigantism for GH and IGF1 and precocious puberty for sex steroids) will lead to increased growth velocity[12]. Patients with an overgrowth syndrome of endocrine origin usually present with growth within the normal range before the start of excessive hormonal secretion, which is typically diagnosed owing to new specific signs (breast or testicular enlargement for precocious puberty; pubic hair in cases of androgen secretion; and goitre, exophthalmia and tachycardia for hyperthyroidism). In Cushing syndrome, excess cortisol secretion leads to rapid weight gain leading to extreme obesity, but growth velocity is dramatically reduced. Such conditions with excessive secretion of GH, cortisol, thyroid hormones or sex steroids are extremely rare during early infancy; however, some cases of acromegalo-gigantism due to GH-secreting pituitary adenomas linked to duplications in Xq26 or mutations in *GPR101* can occur before the age of 2 years[13].

Insulin stimulates fetal growth, as demonstrated in children with insulin resistance (such as in those with Donahue syndrome due to mutations of *INSR*, which encodes the insulin receptor)[14] or those with transient neonatal diabetes mellitus, who present with severe growth restriction and neonatal hyperglycaemia[15]. On the other hand, excessive secretion of insulin during fetal life will lead to macrosomia, with an increased risk of neonatal hypoglycaemia. Hyperinsulinism is mainly a result of gestational diabetes mellitus, but some rare genetic conditions can lead to an increase in the production of fetal insulin as in BWS, or in monogenic forms of congenital hyperinsulinism due to mutations in key genes that are implicated in the regulation of insulin secretion by the pancreatic β-cells[16].

Aside from increased levels of some hormones, monogenic obesity typically leads to precocious excessive weight gain, but birth parameters are usually normal. In these rare conditions, in contrast to Cushing syndrome, growth velocity is usually increased but final height is within the normal range. These conditions are commonly caused by mutations in factors that control food intake and satiety[17].

BWS and isolated lateralized overgrowth

Clinical aspects. BWS (MIM #130650) is the most frequent overgrowth syndrome. The prevalence is estimated to be approximately 1 in every 10,500 births[18], but this could be an underestimate because of the existence of incomplete phenotypes. Isolated lateralized overgrowth (ILO)[18] (previously called isolated hemihyperplasia; MIM #235000) was initially defined as a specific condition; however, ILO and BWS are now considered to be part of the Beckwith–Wiedemann spectrum as they share common molecular mechanisms (the definition of Beckwith–Wiedemann spectrum is provided below)[18].

BWS was first reported in the 1960s by John Bruce Beckwith, who described fetuses with overgrowth, exomphalos and adrenal cytomegaly[19], and Hans-Rudolf Wiedemann, who described children with an association of exomphalos, macroglossia and gigantism (termed EMG syndrome)[20]. Aside from these cardinal features (macrosomia, macroglossia, exomphalos and lateralized overgrowth), children with BWS usually present with a facial gestalt, including midface hypoplasia, infraorbital creases and prominent mandible,

Macrosomia
Fetal macrosomia has been defined in several different ways, including birthweight of 4,000–4,500 g (8 lb 13 oz to 9 lb 15 oz) or >90th percentile for gestational age after correcting for neonatal sex and ethnicity. On the basis of these definitions, macrosomia affects 1–10% of all pregnancies. A diagnosis of fetal macrosomia can be made only by measuring birthweight after delivery.

Exomphalos
A midline anterior incomplete closure of the abdominal wall in which there is herniation of the abdominal viscera into the base of the abdominal cord (also known as omphalocele).

Macroglossia
Increased length and width of the tongue.

Lateralized overgrowth
Overgrowth of only one side of the body (also known as hemihypertrophy).

Naevus flammeus
A congenital vascular malformation consisting of superficial and deep dilated capillaries in the skin that result in a reddish to purplish discolouration of the skin.

Visceromegaly
Enlargement of the internal organs in the abdomen, including the liver, spleen, stomach, kidneys or pancreas.

Uniparental disomy
(UPD). The inheritance of two homologous chromosomes from the same parent. These genetic anomalies arise from errors in meiosis and/or mitosis and can occur independently or in combination.

ear creases or pits and facial naevus flammeus[21]. BWS has been associated with many other congenital anomalies, such as hypoglycaemia and hyperinsulinism, cardiac or nephro-urological malformations, cleft palate and polyhydramnios[21]. Furthermore, one study reported an increased prevalence of pre-eclampsia in women during pregnancies in which the child had BWS[22].

Several scoring systems have been proposed to define BWS, each with varying sensitivity and/or specificity concerning the identification of molecular defects[23,24]. These scoring systems are commonly based on the main symptoms, including macroglossia, macrosomia and exomphalos, and some of the other symptoms associated with BWS, such as visceromegaly, lateralized overgrowth, hypoglycaemia or tumours; however, some patients with a molecular defect identified present with only some of the main symptoms. Thus, owing to the high risk of tumours that is associated with this condition, the aim of a clinical scoring system is to avoid patients being given false-negative result (that is, patients carrying a molecular defect who will not be tested for molecular anomalies because of a negative clinical score). To combat the issues with some of the scoring systems, a new scoring system has been defined that includes cardinal features (the presence of these features scores two points) and suggestive features (the presence of these features scores one point)[23] (TABLE 1). In this scoring system, features are considered to be cardinal when frequently observed and specific to BWS. The other criteria are considered to be suggestive because they are not as frequent or are less specific. For example, macroglossia and lateralized overgrowth are considered to be cardinal features, and macrosomia at birth and the occurrence of an embryonic tumour are considered to be suggestive. The purpose of the total score is to define patients for whom a molecular test should be indicated (with a threshold set at two points) and patients with a clinical diagnosis of classic BWS (with a threshold set at four points), whether a molecular defect has been identified or not. In addition to classic BWS, the Beckwith–Wiedemann spectrum includes both patients who fulfill the clinical criteria for classic BWS (irrespective of the identification of a molecular defect) and those who do not but for whom an 11p15 molecular defect has been detected[23]. With regard to the clinical management of patients with Beckwith–Wiedemann spectrum, an international expert consensus group has established 50 recommendations that were published in 2018 (REF.[23]).

Table 1 | **Consensus scoring system proposed for Beckwith–Wiedemann syndrome**

Cardinal features[a]	Suggestive features[b]
Clinical findings	
Macroglossia	Birthweight ≥2 s.d. above the mean
Exomphalos	Umbilical hernia or diastasis recti
Lateralized overgrowth	Facial naevus simplex
Hyperinsulinism that has lasted >1 week and required escalated treatment	Polyhydramnios or placentomegaly
	Ear creases or pits
	Transient hypoglycaemia that has lasted <1 week
	Nephromegaly and/or hepatomegaly
Tumours	
Multifocal and/or bilateral Wilms tumour or nephroblastomatosis	Neuroblastoma, rhabdomyosarcoma, unilateral Wilms tumour, hepatoblastoma, adrenocortical carcinoma or pheochromocytoma
Pathology findings	
Pancreatic adenomatosis, placental mesenchymal dysplasia and adrenal cortex cytomegaly	N/A

From REF.[24]. N/A, not applicable. [a]Cardinal features are scored two points each. [b]Suggestive features are scored one point each. A clinical score of at least two points requires a molecular study of the 11p15 region. A clinical score of four points or more defines a clinical diagnosis of Beckwith–Wiedemann syndrome.

Molecular mechanisms. Approximately 80% of patients with a clinical diagnosis of BWS have a molecular defect within the 11p15 region (FIG. 1). This region includes imprinted genes, which are genes that exhibit monoallelic and parent-of-origin-specific expression. The telomeric domain of 11p15 contains the *IGF2* gene, which promotes fetal growth and is expressed only from the paternal allele. The monoallelic expression of *IGF2* is controlled by an imprinting centre called *H19*/*IGF2*:IG-DMR (where DMR is differentially methylated region) (or imprinting centre 1 (IC1)), which is methylated on the paternal allele only. The centromeric domain of 11p15 contains the *CDKN1C* gene (a cell-cycle-inhibiting factor), which is expressed only from the maternal allele. Expression of *CDKN1C* is controlled by an imprinting centre called *KCNQ1OT1*:TSS-DMR (or IC2), which is methylated on the maternal allele only[25].

Approximately 60% of patients with BWS show abnormal methylation at either IC1 (gain of methylation (GOM) on the maternal allele; IC1 GOM) (5–10%) or IC2 LOM on the maternal allele (50%). Paternal segmental uniparental disomy (UPD) of 11p15 (known as UPD(11)pat)) is observed in approximately 20% of patients with BWS. Loss-of-function mutations in *CDKN1C* are observed in 5–10% of patients with BWS but represent the most frequent mechanism in familial cases of BWS[21,23]. Rare rearrangements of the 11p15 region, such as paternal duplications, have been reported[26]. Finally, a few patients have been reported with whole-genome paternal UPD, for which one study found that patients had a high risk of developing tumours, even in adulthood[27].

Epigenetic defects and UPD are usually diagnosed as somatic events in a mosaic state, whereas mutations in *CDKN1C* or chromosomal rearrangements usually occur as germinal events[28]. The recurrence risk (that is, the risk that BWS will occur again in other family members) is low in cases of an epigenetic defect or UPD. On the other hand, the recurrence risk can be as high as 50% in cases of an inherited mutation in *CDKN1C* or 11p duplication, depending on the sex of the individual who transmits these genetic traits[23]. For example, because of the expression of *IGF2* from the paternal allele, duplications including *IGF2* will lead to BWS only if located on the paternal allele. By contrast, as *CDKN1C* is expressed from the maternal allele, loss-of-function mutations of *CDKN1C* will lead to BWS only if sited on the maternal allele[23]. In 2016, genetic defects that

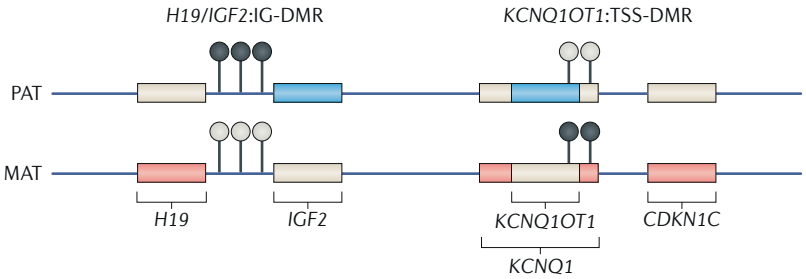

Fig. 1 | **Representation of the 11p15 region in humans.** Blue boxes represent paternally expressed genes and red boxes represent maternally expressed genes. The grey boxes are the silenced alleles. The black and white 'lollipops' represent methylated and unmethylated differentially methylated regions (DMRs), respectively. MAT, maternal allele; PAT, paternal allele.

underlie GOM or LOM were reported within imprinting centres. Approximately 20% of patients with IC1 GOM carry a mutation or deletion in the OCT4 or SOX2 binding sites within IC1. OCT4 and SOX2 are pluripotency factors that are necessary for the protection of maternal IC1 from de novo methylation after fertilization[10,29]. The recurrence risk in siblings can be up to 50% in cases of deletion or mutation within imprinting centres when transmitted by the mother[29,30].

Clinicians can suspect the presence of the Beckwith–Wiedemann spectrum in a fetus during pregnancy, especially in cases of exomphalos, macroglossia or visceromegaly detected by routine ultrasonography monitoring[31]. Furthermore, methylation studies of 11p15 in amniotic fluid are sometimes indicated to distinguish between BWS and other conditions with more severe complications[9]. However, prenatal studies can lead to false-negative results because of the usual mosaicism observed in Beckwith–Wiedemann spectrum, especially if the rate of mosaicism is low[9]. Therefore, clinical and molecular geneticists should be aware of such a possibility, and prenatal molecular testing should take into account the benefit of a positive prenatal diagnosis of BWS versus the possible complications of the prenatal sampling of amniotic fluid and the possibility of a false-negative result.

Approximately 25% of patients with BWS carry epigenetic defects at other imprinted loci in addition to the 11p15 locus. These have been called multilocus imprinting disturbances (MLIDs) and have been observed in other imprinting disorders, such as Silver–Russell syndrome[32,33], Temple syndrome[34], transient neonatal diabetes mellitus[35] and pseudo-hypoparathyroidism[36,37]. The involvement of MLIDs in the clinical presentation of the patients is still unclear, but several studies suggest a more severe phenotype (especially in terms of cognitive development) in patients with MLIDs than in patients without[38]. Since 2015, mutations in the NOD-, LRR- and pyrin domain-containing (*NLRP*) genes have been identified in mothers with recurrent miscarriages and children with imprinting disorders, including patients with BWS[39,40]. The prevalence of such mutations in mothers of patients with imprinting disorders, however, is unknown, but in our opinion such events probably occur rarely. Therefore, screening for mutations in the mothers is not currently recommended as a routine diagnostic procedure when a patient has a molecular diagnosis of BWS[23].

Assisted reproductive technologies. The mechanisms that lead to epigenetic defects are generally unknown (apart from rare deletions or mutations within the imprinting centres or mutations in *NLRP*), giving rise to the hypothesis of an environmental mechanism. The link between imprinting disorders and assisted reproductive technologies was suggested in the 2000s following reports that showed that the parents of patients with BWS or Angelman syndrome were likely to have used assisted reproductive technology for successful conception[41–43]. Intriguingly, patients with Angelman syndrome conceived after assisted reproductive technology often have an LOM at the DMR of the Prader–Willi and Angelman syndromes locus of chromosome 15q11-q13, whereas LOM is observed in <5% of patients with Angelman syndrome who were conceived without the use of assisted reproductive technologies[44,45]. For BWS, in 2017 one group determined a relative risk of 10.7 for children being born with the use of assisted reproductive technology[46], and a meta-analysis found a relative risk of 5.8 (REF.[45]). Most patients with BWS conceived after assisted reproductive technology show IC2 LOM, whereas this mechanism occurs in only 50% of patients with BWS who were conceived without the use of assisted reproductive technologies[41]. To date, no specific technology or aetiology of infertility has been shown to be involved in the occurrence of epigenetic defects, and further studies are needed to decipher the mechanisms that link subfertility, assisted reproductive technologies and imprinting defects.

Simpson–Golabi–Behmel syndrome

Clinical aspects. Simpson–Golabi–Behmel syndrome (SGBS; MIM #312870) is a rare X-linked disorder that was first reported by Joe Leigh Simpson and colleagues[47] and subsequently described by Mahin Golabi, Linda Rosen, Annemarie Behmel and colleagues[48,49]. Since the first descriptions of SGBS, there have been a number of case reports but few clinical reviews[50–52], and the exact prevalence of the disorder has not been precisely evaluated. In one review, the authors noted that 250 patients had been reported in the literature, but they included patients for whom misdiagnosis was possible as no molecular analysis was performed to confirm the cases. However, it is possible that SGBS could still be underdiagnosed as many clinicians are unfamiliar with the phenotype.

In 2013, one group reported the clinical description of 42 male patients and reviewed 63 published cases that had all been confirmed by molecular analysis[51]. For this Review, we have included data made available since the 2013 review, including 18 additional patients tested at our two laboratories (M.-P. Moizard and F.B., unpublished observations) and 29 new descriptions in the literature. This has led to the identification of 152 male individuals with a known mutation, for whom the frequency of the clinical features was estimated.

SGBS has a recognizable clinical picture, and obtaining the correct diagnosis should not be an issue in most cases.

Assisted reproductive technologies
Consist of procedures that involve the in vitro handling of both human oocytes and sperm, or of embryos, with the objective of establishing a pregnancy.

Overgrowth is usually detected prenatally and is often associated with polyhydramnios. Macrosomia at birth is the most frequent finding (86%) and is often associated with macroglossia (78%) and visceromegaly (nephromegaly in 61% and hepatomegaly in 46% of cases), whereas postnatal overgrowth occurs in only slightly more than half of the patients (58%). Similarly, macrocephaly is present in more than half of the patients at birth (57.5%) but less often during postnatal life (43%). Therefore, a height or occipitofrontal circumference (OFC) in the normal range in adulthood does not exclude the diagnosis of SGBS. Patients with SGBS generally have a weight that is appropriate for their stature. They do not seem to be at a high risk of neonatal hypoglycaemia, as this complication has been reported in only eight patients. In most cases (95%), patients with SGBS have a particular facial appearance that can be very similar to young patients with BWS; however, patients with SGBS are distinguishable by the presence of a midline groove of the tongue or lower lip, and ear pits or grooves are less frequent (17%) than in patients with BWS. In addition, supernumerary nipples are frequent (59%) and hand anomalies (broad and/or short hands, brachydactyly, mild cutaneous finger syndactyly and nail dysplasia of the index finger) are suggestive of the condition[53].

Among overgrowth syndromes, SGBS is distinct as it includes a constellation of congenital malformations, among which genitourinary malformations are the most frequent (73%) and diaphragmatic hernia (30%) are the most suggestive. Umbilical hernia and/or diastasis recti, renal dysplasia and heart defects are each observed in approximately one-third of the patients, and cleft lip and/or palate is observed in approximately one-quarter of patients. It is noteworthy that exomphalos has never been reported. Skeletal anomalies are also frequently observed (50%), including chest deformity (pectus excavatum), which is the most common, and rib and vertebral body anomalies, whereas postaxial polydactyly of the hands is infrequent (15%), although it is considered to be a hallmark of the syndrome[50]. As is the case with BWS, neonatal hypotonia and a delay in motor and language development are possible in patients with SGBS, but in our opinion intellectual disability is probably rarer than mentioned in the literature, although no precise study has been performed on the subject. Of note, however, speech problems, accentuated by a cleft palate and/or macroglossia, are frequent, and many patients experience difficulties in school.

There is increasing evidence that some female carriers of a risk allele are symptomatic, usually to a lesser degree than males with the syndrome, but there are no statistical data available in the literature[54]. Large-scale X inactivation studies to understand the underlying mechanism of this phenotypic expression have not yet been reported.

Molecular mechanisms. SGBS is caused by mutations in *GPC3* (which maps to Xq26)[55,56]. This gene encodes GPC3, a 70 kDa core protein of 580 amino acids. GPC3 is one of the six known mammalian glypicans that share a heparan sulfate glycan chain and regulate WNT, Hedgehog, fibroblast growth factor and bone morphogenetic protein signalling[57,58]. GPC3 itself negatively regulates cell proliferation by inhibiting Hedgehog[57] and modulating WNT signalling pathways[58]. In 2018, a review of the molecular data showed that most of the 86 distinct *GPC3* mutations identified to date are unique and 82% are inherited[56]. Of the mutations in *GPC3*, 43% are large rearrangements, with most of these being deletions, followed by truncating point mutations (frameshift or nonsense mutations) dispersed throughout the entire gene, and the mutations are predicted to result in a loss of function[56]. Missense mutations are rare, and the two that were functionally characterized impaired GPC3 function by preventing GPC3 cleavage or transport to the cell surface[59,60]. To date, no genotype–phenotype correlation has been identified.

Other syndromes with general overgrowth

In addition to BWS and SGBS, other syndromes with generalized overgrowth have been described, such as Sotos, Weaver and Perlman syndromes or the more recently described Malan syndrome. These syndromes usually include abnormal intellectual development (refer to the description of these syndromes thereafter). Many of these syndromes are caused by genetic defects in genes that are involved in the regulation of epigenetic markers, such as DNA methylation or histone modification[4]. Most of these genes are also altered as somatic events in cancers, which reinforces the hypothesis that alteration of the expression and/or activity of factors that control physiological cell proliferation can lead to either abnormal growth (overgrowth or growth retardation) or cancer.

Sotos syndrome (MIM #117550) is a cause of overgrowth syndrome described first by Juan Fernandez Sotos in 1964 (REF.[61]). Patients usually present with excessive birth length (whereas birthweight is typically not affected), excessive postnatal growth and advanced bone age[62,63]. The OFC is usually increased at all ages. Jaundice, hypotonia and reduced feeding are frequent in neonates[62,63]. A delay in achieving early developmental milestones, particularly motor skills, is common.

Most patients with Sotos syndrome have some degree of intellectual impairment, ranging from mild to severe learning disability. Cognitive development is usually more impaired in patients with Sotos syndrome than in patients with BWS or SGBS, with learning disabilities frequently being reported in patients[64]. Furthermore, patients often exhibit behavioural problems and symptoms of autism spectrum disorder[65]. Up to 40% of patients experience seizures[62,66]. Patients often present with typical facies, with a long face, large forehead with sparse frontotemporal hair, down-slanting palpebral fissures, malar flushing and a typical long and prominent chin[62,67]. Sotos syndrome can be associated with several malformations, including those of the heart, kidney and brain[62]. Skeletal signs can also be present, mainly scoliosis (up to 50%) but also flat feet and genu varum or genu valgum, which is possibly linked to the hyperlaxity that is commonly observed[63].

Sotos syndrome is mainly caused by mutations or deletions of *NSD1* (REF.[68]). *NSD1* encodes a histone-methyltransferase protein (methylation of H3K36) and

Diastasis recti
A separation of the rectus abdominis muscle into right and left halves (which are normally joined at the midline at the linea alba).

Pectus excavatum
A defect of the chest wall characterized by a depression of the sternum, giving the chest (pectus) a caved-in (excavatum) appearance.

Postaxial polydactyly
A form of polydactyly in which the extra digit or digits are localized on the side of the fifth finger or fifth toe.

Genu varum
A positional abnormality marked by outward bowing of the legs in which the knees stay wide apart when a person stands with the feet and ankles together.

Genu valgum
A positional abnormality in which the legs angle inward, such that the knees are close together and the ankles are far apart.

Microretrognathism
A form of developmental hypoplasia of the mandible in which the mandible is mislocalized posteriorly.

is therefore involved in the control of epigenetic markers and gene transcription. These mutations usually occur as a de novo event[69]. Mutations in *SETD2*, *DNMT3A* or *APC2* have also been described in patients with a clinical presentation close to Sotos syndrome[70–72]. *SETD2* encodes a histone-methyltransferase protein controlling the methylation of H3K36, just as is the case with NSD1 (REF.[73]). *DNMT3A* encodes a DNA methyltransferase that can bind to H3K4me0 and thus is also involved in transcriptional control[74]. To date, three individuals with mutations in *SETD2* and a Sotos phenotype have been described in the literature[70,71]. Mutations in *SETD2* have also been described in patients with an autism spectrum disorder; these data are part of studies investigating large cohorts of individuals with an autism spectrum disorder using next-generation sequencing[75–77]. One mutation of *APC2* has been reported in two siblings with overgrowth and intellectual disability and a phenotype compatible with Sotos syndrome[72]. APC2 is involved in brain development and is a downstream target of NSD1. Interestingly, APC2-deficient mice show a large OFC, cerebral anomalies and abnormal behaviour but no overgrowth[72].

In comparison, 27 patients with a Sotos-like phenotype have been reported to harbour mutations in *DNMT3A*[71,78–80]; however, patients with mutations in *DNMT3A* are now reported to have Tatton–Brown–Rahman syndrome (TBRS; MIM #615879), a condition that overlaps with Sotos syndrome, associating overgrowth, obesity, intellectual disability, kyphoscoliosis, seizures and frequent psychiatric issues[81]. Patients with Sotos-like syndrome and mutations in *SETD2* have facial signs highly reminiscent of Sotos syndrome, whereas patients with TBRS have a quite different facial morphology (including low-set, horizontal, thick eyebrows; narrow palpebral fissures; coarse features; round face; and enlargement of the two upper central incisors) even if the other symptoms clearly mimic Sotos syndrome[78,81].

In 2010, deletions or mutations in the *NFIX* gene were identified in patients with a Sotos-like phenotype, which has been referred to as Malan syndrome (MIM #614753)[82]. Patients with Malan syndrome often have a slightly increased length and OFC at birth and a facial aspect close to that of patients with Sotos syndrome. Ocular abnormalities, pectus excavatum and scoliosis have been reported in patients with Malan syndrome[83,84], defining an intermediate phenotype between Sotos syndrome and Marfan syndrome (MIM #154700; a disorder of connective tissue, with ocular, skeletal and cardiovascular manifestations and tall stature)[85]. Learning disabilities are almost universal in patients with Malan syndrome and can be anything from moderate to severe[83]. Mutations in *NFIX* have also been identified in Marshall–Smith syndrome (MIM #602535), a rare condition with skeletal dysplasia, psychomotor delay, failure to thrive, respiratory distress and facial dysmorphism (including high forehead, underdeveloped midface, anteverted nares and retrognathism)[86]. This different phenotype might be explained by different types or locations of mutations. Mutations or deletions of *NFIX* leading to haploinsufficiency or loss of the ability to bind DNA lead to Malan syndrome, whereas mutations with a dominant-negative effect lead to Marshall–Smith syndrome[84].

The growth phenotype in patients with Weaver syndrome (MIM #277590) is usually similar to that of patients with Sotos syndrome, with a high birth length and large OFC, a tall postnatal stature and advanced bone age. However, the facial gestalt is usually different, and patients have large fleshy ears and specificity concerning the chin, as patients with Weaver syndrome often have microretrognathism and a horizontal crease of the chin[87]. Other clinical features observed in patients with Weaver syndrome are almond-shaped palpebral fissures, widely spaced eyes and a broad forehead (with the phenotype becoming less evident with age) in addition to umbilical hernia and soft doughy skin[87]. Suggestive features are camptodactyly of the fingers and toes[87]. Bone age is often greatly advanced (even more so than in patients with Sotos syndrome) but without advanced tooth eruption. Patients with Weaver syndrome can have poor coordination, abnormal tone and a hoarse low cry in infancy[87]. Cognitive development is usually impaired in patients with Weaver syndrome, but intellect varies widely, from nearly within the normal range to severely impaired[87]. Weaver syndrome is caused by mutations within *EZH2*, which encodes a histone methyltransferase and is therefore associated with the regulation of gene transcription[88]. Furthermore, whole-exome sequencing allowed the identification of mutations in *EED* (a cofactor of EZH2) in patients with a Weaver-like phenotype[89–91].

Perlman syndrome (MIM #267000) was first described in the 1970–1980s[92]. Children with Perlman syndrome have a phenotype close to that of BWS, with fetal overgrowth, but visceromegaly (and especially nephromegaly) is usually very prominent. Affected children are usually hypotonic, with neurodevelopmental delay, and have facial dysmorphism (prominent forehead, broad and flat nasal bridge, inverted V-shaped upper lip and low-set ears)[93]. Mortality is high in newborn babies owing to renal dysplasia, and more than half of the children who survive after birth will develop Wilms tumour[93]. Homozygous mutations in *DIS3L2* were identified in children with Perlman syndrome in 2012 (REF.[94]). *DIS3L2* encodes an exoribonuclease, which has a major role in controlling the degradation of a number of coding and non-coding RNAs[95]. DIS3L2 also has a role in the regulation of mitosis and cellular proliferation, as the protein is also involved in the exosome machinery.

PTEN mutation-related syndromes

PTEN is a key negative regulator of the PI3K–AKT–mTOR signalling pathway[96] (for more information on this pathway, see the section on syndromes with segmental overgrowth later in the manuscript). Patients harbouring constitutional mutations of *PTEN* can present with various phenotypes, which have been grouped into the PTEN hamartoma tumour syndrome[97]. This syndrome includes Cowden syndrome (MIM #158350) and Bannayan–Riley–Ruvalcaba syndrome (MIM #153480). Gastrointestinal hamartomatous polyposis, breast cancer, mucocutaneous papillomatous papules

and penile freckling associated with vascular or lymphatic malformations are very frequent in patients with these syndromes. With regard to growth, patients usually have macrocephaly but stature within the normal range[97]. Developmental delay and/or autism spectrum disorders can be observed, particularly in Bannayan–Riley–Ruvalcaba syndrome, for which intellectual disability is observed in 50% of patients[97].

Syndromes with segmental overgrowth

In addition to BWS, several pathological conditions include segmental overgrowth, including congenital lipomatous overgrowth with vascular, epidermal and skeletal anomalies (CLOVES; MIM #612918); megalencephaly–capillary malformation (MCAP; MIM #602501); Klippel–Trenaunay syndrome (MIM #149000); and others (such as fibroadipose hyperplasia and macrodactyly)[98]. All of these syndromes with segmental overgrowth are grouped under the term 'PIK3CA-related overgrowth syndromes' or PROS. Segmental overgrowth syndromes also include Proteus syndrome (MIM #176920) or hypoinsulinaemic hypoglycaemia with hemihypertrophy (HIHGHH). These two latter syndromes usually include cutaneous and/or vascular malformations, which lead to segmental overgrowth of part of the body[99].

Mutations (which usually lead to a gain of function of the protein) in oncogenic pathways have been identified in these syndromes. The PI3K–AKT–mTOR signalling pathway has been implicated in tumorigenesis and is therefore a target for cancer therapy[96]. Mutations in *PIK3CA* were initially described in Klippel–Trenaunay syndrome, a condition that is associated with capillary and vascular malformations as well as overgrowth. The spectrum of *PIK3CA* mutations has now been broadened, as somatic mutations in *PIK3CA* have been identified in CLOVES, MCAP, fibroadipose hyperplasia and hemimegalencephaly[98,100]. In addition to segmental overgrowth, these rare conditions are characterized by vascular, cerebral, cutaneous and skeletal anomalies[98,100]. Depending on the presence of such anomalies, criteria for *PIK3CA* molecular testing have been proposed. Furthermore, criteria for a diagnosis of PROS have been proposed, including the presence of a *PIK3CA* mutation[96,100].

The same mutation hot spots in *PIK3CA* have been identified in either syndromes with segmental overgrowth or cancer (for example, p.Glu542Ly, p.Glu545Lys and p.His1047Arg). Mutations in patients with PROS are usually observed in a mosaic state and might therefore be undetectable in circulating blood cells and be observed only in hyperplastic tissues (such as bone, skin, fatty tissue, nerves or vessels) with variable rates of mosaicism[86,98,101]. Deep-targeted next-generation sequencing approaches are highly performant tools to detect such somatic mutations, especially for mutations with low mosaicism undetectable by Sanger sequencing[86,102]. Constitutional *PIK3CA* mutations (that is non-mosaic mutations that are detectable in circulating blood cells) have also been reported. In the latter case, generalized overgrowth is usually observed, including diffuse megalencephaly, with some symptoms that can overlap with those of BWS, such as exomphalos, hypoglycaemias and visceromegaly[102].

Germline mutations of *PTEN* have been identified in patients with several conditions that include overgrowth (Cowden syndrome and Bannayan–Riley–Ruvalcaba syndrome; see earlier in manuscript for more information on these)[97]. Somatic loss-of-function mutations of *PTEN* have been identified in many types of tumour[96]. Mosaic mutations of *PTEN* have now also been identified in some cases of segmental overgrowth[103].

Proteus syndrome has been described in patients with segmental overgrowth with a lipomatous cerebriform aspect of the hyperplastic tissues. In Proteus syndrome, segmental overgrowth is usually absent or barely detectable at birth and develops progressively after birth. Aside from overgrowth, additional clinical criteria for a positive diagnosis of Proteus syndrome have been proposed, including cutaneous naevi, vascular or lymphatic anomalies or abnormal adipose tissue (hyperplasia or lipoatrophy)[99]. A few patients have also been described with HIHGHH, which includes segmental overgrowth, but this is rare. Proteus syndrome and HIHGHH have been linked to mutations in *AKT1* (REF.[104]) and *AKT2* (REF.[105]). The involvement of germline mutations of *PTEN* in Proteus syndrome is uncertain, as some patients carrying *PTEN* mutations might have been misdiagnosed as having Proteus syndrome[106,107].

Mutations of other members of the PI3K–AKT–mTOR pathway (*AKT3* (REF.[108]), *CCND2* (REF.[109]) and *PIK3R2* (REF.[108])) have been identified in rare conditions that include megalencephaly polymicrogyria polydactyly hydrocephalus syndrome, reinforcing the predominant role of the PI3K–AKT–mTOR pathway in the control of tissue growth and cerebral development.

Tumour risk and tumour screening

Overgrowth syndromes are usually associated with an increased risk of tumours (TABLE 2); however, the prevalence of tumours in most overgrowth syndromes is only slightly increased compared with the general population, unlike other conditions, such as Li–Fraumeni syndrome (linked to mutations in *TP53*) or mutations in *BRCA1* and *BRCA2*, which have a nearly complete penetrance of tumour development[110,111]. International consensus statements are needed to assess the specific issue of the indication and modality of tumour screening in patients with overgrowth syndromes.

The association between overgrowth syndromes and increased tumour risk has been particularly well described in patients with BWS. Sporadic (that is, not occurring in a syndromic context) embryonic tumours, such as Wilms tumours, adrenocortical carcinomas and hepatoblastomas, all overexpress *IGF2* (REFS[112–114]). Furthermore, molecular defects in the 11p15 region are frequently observed in sporadic Wilms tumours and adrenocortical carcinomas (loss of heterozygosity or imprinting)[113,115,116]. In patients with BWS, the overall tumour risk is estimated to be approximately 7%, but the prevalence of tumours is extremely variable and depends on the molecular mechanism that causes BWS. Indeed, the prevalence could be as high as 20% in patients with IC1 GOM and 12% in patients with

Hemimegalencephaly
Enlargement of all or parts of one cerebral hemisphere.

Polymicrogyria
A congenital abnormality of the cerebral hemisphere characterized by an excessive number of small gyri (convolutions) on the surface of the brain.

Table 2 | **Reported recommendations for tumour screening in overgrowth syndromes**

Syndrome	Prevalence (%)	Type of tumour	Screening programme	Refs
Recommendations based on consensus meetings				
Beckwith–Wiedemann syndrome	7[a]	Wilms tumour	Abdominal ultrasonography every 3 months until 7 years of age for the high-risk groups[a]	24
		Hepatoblastoma		
		Neuroblastoma		
Sotos syndrome	3	Neuroblastoma	No screening	120
		Teratoma		
Weaver syndrome	Unknown	Hodgkin disease	No screening	120
		Acute lymphoid leukaemia		
		Neuroblastoma		
PTEN-related hamartoma tumour syndrome	Up to 75	Thyroid	Thyroid ultrasonography from 7 years of age	127,128
		Breast		
		Kidney	Screening for melanoma and breast, endometrial and colorectal carcinomas once an adult (>18 years of age)	
		Endometrial carcinoma		
Recommendations with no consensus				
Simpson–Golabi–Behmel syndrome	8	Wilms tumour	Abdominal ultrasonography every 3 months until 7 years of age	50,121
		Neuroblastoma	Urine catecholamines	
		Hepatoblastoma	Serum α-fetoprotein	
Perlman syndrome	Up to 40	Wilms tumour	Abdominal ultrasonography every 3 months until 7 years of age	121
PIK3CA mutations	Unknown	Wilms tumour	No screening or abdominal ultrasonography every 3 months until 7 years of age	100
Malan syndrome	Not reported	N/A	No screening	N/A

N/A, not applicable. [a]Depending on the molecular subtype. Note that this programme differs from that of the American Association for Cancer Research[120], which recommends abdominal screening for any patient with Beckwith–Wiedemann syndrome.

UPD(11)pat, whereas the prevalence is approximately 2% in patients with IC2 LOM[32,117,118]. Given the types of embryonic tumour, the prevalence of Wilms tumour is particularly high for patients with IC1 GOM or UPD(11)pat, whereas evidence for Wilms tumour in patients with IC2 LOM or *CDKN1C* mutations is anecdotal[119]. Finally, patients who present with classic BWS but no identified molecular defect also have an increased risk of tumours (especially Wilms tumour)[118]. This observation led to international recommendations for tumour screening that are stratified depending on the molecular aetiology, with abdominal ultrasound scans recommended only for the high-risk groups, which include patients with a clinical diagnosis of BWS and no identified molecular defect[23]. These international recommendations vary from the recommendations of the American Association for Cancer Research (AACR), which recommends screening for all patients, irrespective of the molecular cause, and therefore have a lower threshold to trigger tumour screening[120].

In SGBS, the risk of developing an embryonal tumour has been reported to be 10%, but the reporting study included only three cases (one hepatocarcinoma, one gonadoblastoma and one neuroblastoma), and the authors did not conduct a molecular analysis[121]. We reviewed 152 patients with a *GPC3* mutation and found 1 fetus with nephroblastomatosis, 5 patients with Wilms tumour, 6 with hepatoblastoma and 1 with medulloblastoma (M.-P. Moizard and F.B., unpublished observations). The occurrence of leukaemia in one patient could have been coincidental, as haematological malignancies would not be expected in SGBS given the absence of *GPC3* expression in white blood cells[55]. With leukaemia removed from risk equations, the overall frequency of tumour risk is 8.5%, but the small size of the sample hampers precise determination of the frequency. To date, no consensus has been reached concerning tumour screening in patients with SGBS. Pablo Lapunzina and colleagues suggested that patients with SGBS undergo tumour surveillance with abdominal ultrasonography and measurements of serum levels of α-fetoprotein and urinary levels of catecholamine[121]; however, no evaluation of the effectiveness of this surveillance has been reported. In the absence of a genotype–phenotype correlation, we believe that it could be advisable to perform at least a clinical and abdominal ultrasonography surveillance in patients with SGBS until the age of 7 years, as is done in patients BWS, while we await further studies.

Somatic mutations affecting *NSD1* or *EZH1* have been identified in various types of tumour, suggesting a tumour suppressor role[122,123]. Concerning germline mutations in Sotos and Weaver syndromes, the prevalence of tumours is relatively low (probably <5%); this

includes neuroblastoma, teratoma, acute leukaemia and small-cell lung cancer[87,124]. No tumour screening has been recommended by the AACR for patients with Sotos or Weaver syndromes because of the relatively low risk and varying tumour type[124]. To date, no tumour has been associated with Malan syndrome.

The prevalence of Wilms tumour in patients with Perlman syndrome is very high (up to 64% of patients who survive beyond the neonatal period)[93]. No widely accepted recommendation has been made regarding tumour screening in patients with Perlman syndrome; however, some experts recommend abdominal screening with ultrasonography in children with Perlman syndrome, as is carried out in children with BWS, given the very high prevalence of Wilms tumour[121]. Further studies are needed to determine the optimal age at which such screening should be performed.

Patients with *PTEN* mutations have a very high risk of malignant tumours, especially those of the breast, kidney, thyroid, skin or endometrium, with a penetrance of approximately 75%[125]. These tumours usually occur during adulthood, with the exception of thyroid carcinomas, which can be present during childhood[126]. Given the very high risk of malignant tumours, specific screening protocols have been proposed, including ultrasonography screening for thyroid carcinoma from the age of 7 years[127] and screening for colorectal, breast and endometrial carcinomas during adulthood, in addition to screening for melanomas[128]. Disruption of PTEN or proteins from the AKT family is frequently observed as a somatic event in various types of tumour (mostly carcinomas)[11]. With regard to somatic mutations of *PTEN* or *AKT1* in Proteus syndrome, various types of tumour have been reported, most of which are benign[99]. Screening has not been recommended given the large spectrum of tumours with regard to tumour types and variation in the age of occurrence[99].

Wilms tumour or nephroblastomatosis has been reported in patients with *PIK3CA* mutation-related syndromes[101,129], but the prevalence of tumours associated with *PIK3CA* mutations has not been accurately assessed. Given the reported tumours, however, caution should be advised, and abdominal tumour screening has been suggested[100].

Clinical overlap

Despite specific traits for each syndrome, overgrowth syndromes often share clinical symptoms (TABLE 3). As described before in this Review, this is particularly true for patients with BWS or SGBS who can have macroglossia, macrosomia, umbilical hernia and the same spectrum of embryonic tumours.

Neonatal or postnatal macrosomia is a frequent finding among overgrowth syndromes, as more than half of the patients present with this symptom; however, a gradient can be observed for postnatal growth for Sotos syndrome, SGBS and BWS, with children with Sotos syndrome being the tallest and patients with BWS often being of average or slightly above-average height. OFC can also be used to distinguish between these three syndromes, as patients with Sotos syndrome often have a very large OFC, whereas the OFC is only slightly increased in children with SGBS and is usually within the normal range for children with BWS. Neurocognitive development is often impaired in patients with Sotos syndrome and is usually within the normal range or

Table 3 | **Clinical description of the most common overgrowth syndromes**

Syndrome	MIM	Gene or genes	Severity of overgrowth	Severity of macrocephaly	Severity of cognitive impairment
Generalized overgrowth					
Beckwith–Wiedemann syndrome	#130650	*IGF2/CDKN1C*[a]	+	0	0[b]
Simpson–Golabi–Behmel syndrome	#312870	*GPC3*	+	0/+	0/+
Sotos syndrome	#117550	*NSD1*	++	++	+/++
Weaver syndrome	#277590	*EZH2* and *EEP*	++	++	+/++
Malan syndrome	#614753	*NFIX*	++	++	+/++
Perlman syndrome	#267000	*DIS3L2*	+	0/+	+/++
Segmental overgrowth					
Beckwith–Wiedemann syndrome	#130650	*IGF2*[a] and *CDKN1C*[a]	+	0	0[b]
PTEN-related hamartoma tumour syndrome	#158350 and #153480	*PTEN*	+/++	++	+/++
PIK3CA-related overgrowth syndrome	#612918, #149000 and #602501	*PIK3CA*	+/++	++	+/++
Proteus syndrome	#176920	*AKT1* and *PTEN*	+/++	0	0

0, phenotype absent; +, mild phenotype; ++, severe phenotype; +/++ mild to severe phenotype. [a]*IGF2* and *CDKN1C* are two imprinted genes mapped at 11p15.5. Abnormal methylation at imprinting centres within 11p15.5 (imprinting centre 1 or 2) represents the main molecular mechanism of Beckwith–Wiedemann syndrome (see main text). [b]At the exclusion of patients with severe neonatal complications (prematurity or hypoglycaemia).

	Overgrowth				
Type of overgrowth	Generalized overgrowth			Segmental overgrowth	
Additional features	Large OFC, mental retardation and seizures	Macroglossia		Lipomatosis and cerebriform connective tissue naevi	Large OFC, vascular or capillary malformations and cutaneous, skeletal and cerebral abnormalities
		Supernumerary nipples and polydactyly	Omphalocele		
Clinical diagnosis	Sotos syndrome, Weaver syndrome and Malan syndrome	Simpson–Golabi–Behmel syndrome	Beckwith–Wiedemann spectrum	Proteus syndrome	PIK3CA-related overgrowth spectrum
First-step molecular test	Sanger sequencing or NGS (For NGS, a multiple-gene panel for overgrowth syndromes is preferred. Patients can present with distinguishable phenotypes; thus, a candidate gene approach can be indicated if single-gene Sanger sequencing is used)		Methylation studies at 11p15 (consider *CDKN1C* sequencing depending on the family history)	NGS on affected tissue	
Molecular defect	Mutation or deletion in *NSD1*, *EZH2* or *NFIX*	Mutation or deletion in *GPC3*	IC1 GOM, IC2 LOM and UPD(11)pat	Mutation in *PTEN* or *AKT1*	Mutation in *PIK3CA*
Second-step molecular test	Consider: • Multiplex ligation-dependent probe amplification or SNP or CGH array • 11p15 methylation studies or exome sequencing		NGS	Methylation studies at 11p15 on alternative tissue	NGS on alternative tissue

Fig. 2 | **Proposed molecular testing strategy for overgrowth syndromes.** In cases of generalized overgrowth, Beckwith–Wiedemann syndrome, Simpson–Golabi–Behmel syndrome and Sotos or Sotos-like syndromes should be tested for using molecular studies of circulating blood cells. In cases of segmental overgrowth, hyperplastic tissues should first be investigated for molecular anomalies. Next-generation sequencing (NGS) technologies should be advised, with the exception of Beckwith–Wiedemann syndrome, in which methylation studies should be performed first as they will lead to the identification of a molecular defect in 70–75% of patients with Beckwith–Wiedemann syndrome. CGH, comparative genomic hybridization; GOM, gain of methylation; IC1, imprinting centre 1; LOM, loss of methylation; OFC, occipitofrontal circumference; SNP, single-nucleotide polymorphism; UPD(11)pat, paternal uniparental disomy of chromosome 11.

only slightly impaired in patients with SGBS and patients with BWS. Lateralized overgrowth has been described in patients with BWS but not in patients with Sotos syndrome or SGBS; however, lateralized overgrowth can also be observed in patients with mutations in *PTEN*, *PIK3CA* or *AKT1*.

Physicians who are experienced in the clinical diagnosis of overgrowth syndromes can easily distinguish between the different conditions if patients present with a classic presentation. Some patients, however, can present with incomplete or non-classic phenotypes, leading to the definition of some of these disorders as a spectrum instead of a syndrome[23]. Several studies showed that a molecular overlap can be observed between overgrowth syndromes, as some patients with an initial clinical diagnosis of BWS might have mutations in *NSD1*, and patients with an initial clinical diagnosis of Sotos syndrome might have molecular anomalies within the 11p15 region[130,131]. The same observation has been made between BWS and SGBS (C. Gicquel, unpublished observation).

Molecular diagnostic procedures of overgrowth syndromes should look for mutations, methylation defects or chromosome rearrangements. To this purpose, different procedures (based on sequencing analyses, methylation studies or arrays, respectively) can be used, depending on the clinical presentation (FIG. 2). Methylation studies at 11p15 detect methylation defects (IC1 GOM or IC2 LOM) or UPD(11)pat, which confirms BWS, but do not detect mutations in *NSD1*, *GPC3* or *CDKN1C*, which cause Sotos syndrome, SGBS and BWS, respectively. On the other hand, Sanger sequencing or next-generation sequencing based on gene panels or whole-exome sequencing can detect mutations in these genes but cannot detect methylation defects at 11p15. Thus, in cases of suspected BWS (especially if lateralized overgrowth is present), molecular investigations should include methylation studies of 11p15 as these techniques will allow the detection of a molecular defect in approximately 70–75% of patients[28]. If the result of the molecular investigation is negative, re-examination of the clinical presentation of the patients is recommended to consider alternative diagnoses and further analyses[23]. In the presence of lateralized overgrowth, mosaicism should be considered and methylation studies on an alternative tissue should be considered[23].

In the absence of lateralized overgrowth, most laboratories have developed next-generation-sequencing-based approaches to look for mutations in the genes that have been implicated in overgrowth syndromes[4]. If the initial clinical diagnosis was that of SGBS or Sotos syndrome, next-generation sequencing should be considered first as it will detect point mutations and small deletions. If the result of next-generation sequencing is negative, depending on the clinical presentation, physicians should consider either multiplex ligation-dependent probe amplification (which is usually specific for one locus) or array technologies (which can detect rearrangements such as large deletions or duplications) or methylation studies of 11p15 (especially for patients with SGBS).

A diagnosis of BWS is unlikely if the patient shows intellectual disability associated with overgrowth (except

for those with severe perinatal complications, such as severe persistent hypoglycaemia or very preterm birth)[23]. In this case, next-generation sequencing approaches could be performed first as they will lead to the identification of a molecular defect in up to 50% of the patients (the most prevalent one being in *NSD1*)[4].

With regard to segmental overgrowth, distinguishing between the different conditions can sometimes be challenging. Although a diagnosis of MCAP syndrome can be quite simple in cases of segmental overgrowth associated with megalencephaly, the phenotype can be mild in some cases of *PIK3CA* mutations and mimic BWS. For example, patients can present with only lateralized overgrowth and vascular malformations of the face, which are also observed in BWS[86]. If segmental overgrowth is associated with brain malformations, molecular studies should initially include next-generation sequencing of peripheral blood leukocytes and/or hyperplastic tissues to look for mutations in *PIK3CA*, *PTEN*, *AKT1* or *AKT2*, which are usually present in a mosaic state. Thus, ultradeep next-generation sequencing techniques are needed (those able to detect levels of mutant alleles as low as 1%). Such techniques allow identification of a molecular defect in up to 66% of patients, with a much better rate of detection of these anomalies in the hyperplastic tissue than in circulating blood or buccal swab cells[102,132].

Conclusion

Overgrowth syndromes are mainly caused by the epigenetic and genetic disruption of several factors involved in cell proliferation and/or the regulation of gene expression (regulation of epigenetic markers or transcriptional and/or post-transcriptional processes). Anomalies in the same genes and/or pathways that cause overgrowth syndromes are often observed in tumours, which might explain the increased tumour risk in overgrowth syndromes. A clinical overlap is observed between these rare conditions; however, distinguishing between these conditions is necessary to provide the best patient care because of interdifferences in tumour surveillance (which should be stratified depending on the molecular anomaly), the type of molecular test (for example, methylation analysis or gene sequencing of circulating blood cells or hyperplastic tissue) and genetic counselling (which depends on the result of the molecular test). A detailed clinical description of the exact syndrome a patient has is therefore necessary, with particular attention to the OFC and the evaluation of cognitive development.

Advances in molecular biology have increased the frequency of the identification of molecular defects in patients with an overgrowth syndrome, including somatic mutations in syndromes associated with segmental overgrowth. Consensus meetings involving international experts should be established, in collaboration with patient associations, to redefine these conditions (taking into consideration the molecular defects in the recently identified factors) and establish guidelines concerning the molecular diagnosis and clinical management of these rare diseases.

Published online 6 March 2019

1. Mussa, A. et al. (Epi)genotype-phenotype correlations in Beckwith-Wiedemann syndrome. *Eur. J. Hum. Genet.* **24**, 183–190 (2016).
2. Burton, G. J. & Jauniaux, E. Pathophysiology of placental-derived fetal growth restriction. *Am. J. Obstet. Gynecol.* **218**, S745–S761 (2018).
3. Buchanan, T. A., Xiang, A. H. & Page, K. A. Gestational diabetes mellitus: risks and management during and after pregnancy. *Nat. Rev. Endocrinol.* **8**, 639–649 (2012).
4. Tatton-Brown, K. et al. Mutations in epigenetic regulation genes are a major cause of overgrowth with intellectual disability. *Am. J. Hum. Genet.* **100**, 725–736 (2017).
5. Matsuoka, S. et al. p57KIP2, a structurally distinct member of the p21CIP1 Cdk inhibitor family, is a candidate tumor suppressor gene. *Genes Dev.* **9**, 650–662 (1995).
6. Stampone, E. et al. Genetic and epigenetic control of CDKN1C expression: importance in cell commitment and differentiation, tissue homeostasis and human diseases. *Int. J. Mol. Sci.* **19**, E1055 (2018).
7. Giabicani, E., Netchine, I. & Brioude, F. New clinical and molecular insights into Silver-Russell syndrome. *Curr. Opin. Pediatr.* **28**, 529–535 (2016).
8. Arboleda, V. A. et al. Mutations in the PCNA-binding domain of CDKN1C cause IMAGe syndrome. *Nat. Genet.* **44**, 788–792 (2012).
9. Eggermann, T. et al. Prenatal molecular testing for Beckwith-Wiedemann and Silver-Russell syndromes: a challenge for molecular analysis and genetic counseling. *Eur. J. Hum. Genet.* **24**, 784–793 (2016).
10. Abi Habib, W. et al. Genetic disruption of the oncogenic HMGA2-PLAG1-IGF2 pathway causes fetal growth restriction. *Genet. Med.* **20**, 250–258 (2018).
11. Cheung, M. & Testa, J. R. Diverse mechanisms of AKT pathway activation in human malignancy. *Curr. Cancer Drug Targets* **13**, 234–244 (2013).
12. Baron, J. et al. Short and tall stature: a new paradigm emerges. *Nat. Rev. Endocrinol.* **11**, 735–746 (2015).
13. Trivellin, G. et al. Gigantism and acromegaly due to Xq26 microduplications and GPR101 mutation. *N. Engl. J. Med.* **371**, 2363–2374 (2014).
14. Ben Harouch, S., Klar, A. & Falik Zaccai, T. C. INSR-related severe syndromic insulin resistance. *GeneReviews* https://www.ncbi.nlm.nih.gov/books/NBK476444 (updated 25 Jan 2018).
15. Temple, I. K. & Mackay, D. J. G. Diabetes mellitus, 6q24-related transient neonatal. *GeneReviews* https://www.ncbi.nlm.nih.gov/books/NBK1534 (updated 13 Sep 2018).
16. Nessa, A., Rahman, S. A. & Hussain, K. Hyperinsulinemic hypoglycemia - the molecular mechanisms. *Front. Endocrinol. (Lausanne)* **7**, 29 (2016).
17. Albuquerque, D., Stice, E., Rodriguez-Lopez, R., Manco, L. & Nobrega, C. Current review of genetics of human obesity: from molecular mechanisms to an evolutionary perspective. *Mol. Genet. Genomics* **290**, 1191–1221 (2015).
18. Kalish, J. M. et al. Nomenclature and definition in asymmetric regional body overgrowth. *Am. J. Med. Genet. A* **173**, 1735–1738 (2017).
19. Beckwith, J. B. in *Annual Meeting of Western Society of Pediatric Research* (WSPR, Los Angeles, California, 1963).
20. Wiedemann, H. R. The EMG-syndrome: exomphalos, macroglossia, gigantism and disturbed carbohydrate metabolism [German]. *Z. Kinderheilkd* **106**, 171–185 (1969).
21. Shuman, C., Beckwith, J. B. & Weksberg, R. Beckwith-Wiedemann syndrome. GeneReviews https://www.ncbi.nlm.nih.gov/books/NBK1394 (updated 11 Aug 2016).
22. Romanelli, V. et al. CDKN1C mutations in HELLP/preeclamptic mothers of Beckwith-Wiedemann Syndrome (BWS) patients. *Placenta* **30**, 551–554 (2009).
23. Brioude, F. et al. CDKN1C mutation affecting the PCNA-binding domain as a cause of familial Russell Silver syndrome. *J. Med. Genet.* **50**, 823–830 (2013).
24. Brioude, F. et al. Expert consensus document: clinical and molecular diagnosis, screening and management of Beckwith-Wiedemann syndrome: an international consensus statement. *Nat. Rev. Endocrinol.* **14**, 229–249 (2018).
25. Eggermann, T. et al. Imprinting disorders: a group of congenital disorders with overlapping patterns of molecular changes affecting imprinted loci. *Clin. Epigenetics* **7**, 123 (2015).
26. Heide, S. et al. Chromosomal rearrangements in the 11p15 imprinted region: 17 new 11p15.5 duplications with associated phenotypes and putative functional consequences. *J. Med. Genet.* **55**, 205–213 (2018).
27. Kalish, J. M. et al. Clinical features of three girls with mosaic genome-wide paternal uniparental isodisomy. *Am. J. Med. Genet. A* **161A**, 1929–1939 (2013).
28. Eggermann, T. et al. Clinical utility gene card for: Beckwith-Wiedemann Syndrome. *Eur. J. Hum. Genet.* **22**, 435 (2014).
29. Poole, R. L. et al. Beckwith-Wiedemann syndrome caused by maternally inherited mutation of an OCT-binding motif in the IGF2/H19-imprinting control region, ICR1. *Eur. J. Hum. Genet.* **20**, 240–243 (2012).
30. Abi Habib, W. et al. Extensive investigation of the IGF2/H19 imprinting control region reveals novel OCT4/SOX2 binding site defects associated with specific methylation patterns in Beckwith-Wiedemann syndrome. *Hum. Mol. Genet.* **23**, 5763–5773 (2014).
31. Kagan, K. O. et al. Novel fetal and maternal sonographic findings in confirmed cases of Beckwith-Wiedemann syndrome. *Prenat. Diagn.* **35**, 394–399 (2015).
32. Azzi, S. et al. Complex tissue-specific epigenotypes in Russell-Silver Syndrome associated with 11p15 ICR1 hypomethylation. *Hum. Mutat.* **35**, 1211–1220 (2014).
33. Wakeling, E. L. et al. Diagnosis and management of Silver-Russell syndrome: first international consensus statement. *Nat. Rev. Endocrinol.* **13**, 105–124 (2017).
34. Geoffron, S. et al. Chromosome 14q32.2 imprinted region disruption as an alternative molecular diagnosis of Silver-Russell syndrome. *J. Clin. Endocrinol. Metab.* **103**, 2436–2446 (2018).
35. Mackay, D. J. et al. Hypomethylation of multiple imprinted loci in individuals with transient neonatal diabetes is associated with mutations in ZFP57. *Nat. Genet.* **40**, 949–951 (2008).

36. Maupetit-Mehouas, S. et al. Simultaneous hyper- and hypomethylation at imprinted loci in a subset of patients with GNAS epimutations underlies a complex and different mechanism of multilocus methylation defect in pseudohypoparathyroidism type 1b. *Hum. Mutat.* **34**, 1172–1180 (2013).
37. Mantovani, G. et al. Diagnosis and management of pseudohypoparathyroidism and related disorders: first international Consensus Statement. *Nat. Rev. Endocrinol.* **14**, 476–500 (2018).
38. Poole, R. L. et al. Targeted methylation testing of a patient cohort broadens the epigenetic and clinical description of imprinting disorders. *Am. J. Med. Genet. A* **161A**, 2174–2182 (2013).
39. Docherty, L. E. et al. Mutations in NLRP5 are associated with reproductive wastage and multilocus imprinting disorders in humans. *Nat. Commun.* **6**, 8086 (2015).
40. Begemann, M. et al. Maternal variants in NLRP and other maternal effect proteins are associated with multilocus imprinting disturbance in offspring. *J. Med. Genet.* **55**, 497–504 (2018).
41. Niemitz, E. L. & Feinberg, A. P. Epigenetics and assisted reproductive technology: a call for investigation. *Am. J. Hum. Genet.* **74**, 599–609 (2004).
42. Rossignol, S. et al. The epigenetic imprinting defect of patients with Beckwith-Wiedemann syndrome born after assisted reproductive technology is not restricted to the 11p15 region. *J. Med. Genet.* **43**, 902–907 (2006).
43. Maher, E. R. et al. Beckwith-Wiedemann syndrome and assisted reproduction technology (ART). *J. Med. Genet.* **40**, 62–64 (2003).
44. Cox, G. F. et al. Intracytoplasmic sperm injection may increase the risk of imprinting defects. *Am. J. Hum. Genet.* **71**, 162–164 (2002).
45. Cortessis, V. K. et al. Comprehensive meta-analysis reveals association between multiple imprinting disorders and conception by assisted reproductive technology. *J. Assist. Reprod. Genet.* **35**, 943–952 (2018).
46. Mussa, A. et al. Assisted reproductive techniques and risk of Beckwith-Wiedemann syndrome. *Pediatrics* **140**, e20164311 (2017).
47. Simpson, J. L., Landey, S., New, M. & German, J. A previously unrecognized X-linked syndrome of dysmorphia. *Birth Defects Orig. Artic. Ser.* **11**, 18–24 (1975).
48. Behmel, A., Plochl, E. & Rosenkranz, W. A new X-linked dysplasia gigantism syndrome: identical with the Simpson dysplasia syndrome? *Hum. Genet.* **67**, 409–413 (1984).
49. Golabi, M. & Rosen, L. A new X-linked mental retardation-overgrowth syndrome. *Am. J. Med. Genet.* **17**, 345–358 (1984).
50. Sajorda, B. J., Gonzalez-Gandolfi, C. X., Hathaway, E. R. & Kalish, J. M. Simpson-Golabi-Behmel syndrome type 1. *GeneReviews* https://www.ncbi.nlm.nih.gov/books/NBK1219 (updated 29 Nov 2018).
51. Cottereau, E. et al. Phenotypic spectrum of Simpson-Golabi-Behmel syndrome in a series of 42 cases with a mutation in GPC3 and review of the literature. *Am. J. Med. Genet. C Semin. Med. Genet.* **163C**, 92–105 (2013).
52. Tenorio, J. et al. Simpson-Golabi-Behmel syndrome types I and II. *Orphanet J. Rare Dis.* **9**, 138 (2014).
53. Vuillaume, M. L. et al. CUGC for Simpson-Golabi-Behmel syndrome (SGBS). *Eur. J. Hum. Genet.* https://doi.org/10.1038/s41431-019-0339-z (2019).
54. Schirwani, S. et al. Duplications of GPC3 and GPC4 genes in symptomatic demale carriers of Simpson-Golabi-Behmel syndromes type 1. *Eur. J. Med. Genet.* https://doi.org/10.1016/j.ejmg.2018.07.022 (2018).
55. Pilia, G. et al. Mutations in GPC3, a glypican gene, cause the Simpson-Golabi-Behmel overgrowth syndrome. *Nat. Genet.* **12**, 241–247 (1996).
56. Vuillaume, M. L. et al. Mutation update for the GPC3 gene involved in Simpson-Golabi-Behmel syndrome and review of the literature. *Hum. Mutat.* **39**, 790–805 (2018).
57. Capurro, M. I. et al. Glypican-3 inhibits Hedgehog signaling during development by competing with patched for Hedgehog binding. *Dev. Cell* **14**, 700–711 (2008).
58. Filmus, J. & Capurro, M. Glypican-3: a marker and a therapeutic target in hepatocellular carcinoma. *FEBS J.* **280**, 2471–2476 (2013).
59. Shi, W. & Filmus, J. A patient with the Simpson-Golabi-Behmel syndrome displays a loss-of-function point mutation in GPC3 that inhibits the attachment of this proteoglycan to the cell surface. *Am. J. Med. Genet. A* **149A**, 552–554 (2009).
60. Veugelers, M. et al. Mutational analysis of the GPC3/GPC4 glypican gene cluster on Xq26 in patients with Simpson-Golabi-Behmel syndrome: identification of loss-of-function mutations in the GPC3 gene. *Hum. Mol. Genet.* **9**, 1321–1328 (2000).
61. Sotos, J. F., Dodge, P. R., Muirhead, D., Crawford, J. D. & Talbot, N. B. Cerebral gigantism in childhood. a syndrome of excessively rapid growth and acromegalic features and a nonprogressive neurologic disorder. *N. Engl. J. Med.* **271**, 109–116 (1964).
62. Tatton-Brown, K. et al. Genotype-phenotype associations in Sotos syndrome: an analysis of 266 individuals with NSD1 aberrations. *Am. J. Hum. Genet.* **77**, 193–204 (2005).
63. Tatton-Brown, K., Cole, T. R. P. & Rahman, N. Sotos syndrome. *GeneReviews* https://www.ncbi.nlm.nih.gov/books/NBK1479 (updated 19 Nov 2015).
64. Lane, C., Milne, E. & Freeth, M. Cognition and behaviour in Sotos syndrome: a systematic review. *PLOS ONE* **11**, e0149189 (2016).
65. Lane, C., Milne, E. & Freeth, M. Characteristics of autism spectrum disorder in Sotos syndrome. *J. Autism Dev. Disord.* **47**, 135–143 (2017).
66. Nicita, F. et al. Seizures and epilepsy in Sotos syndrome: analysis of 19 caucasian patients with long-term follow-up. *Epilepsia* **53**, e102–e105 (2012).
67. Cole, T. R. & Hughes, H. E. Sotos syndrome: a study of the diagnostic criteria and natural history. *J. Med. Genet.* **31**, 20–32 (1994).
68. Kurotaki, N. et al. Haploinsufficiency of NSD1 causes Sotos syndrome. *Nat. Genet.* **30**, 365–366 (2002).
69. Rayasam, G. V. et al. NSD1 is essential for early post-implantation development and has a catalytically active SET domain. *EMBO J.* **22**, 3153–3163 (2003).
70. Luscan, A. et al. Mutations in SETD2 cause a novel overgrowth condition. *J. Med. Genet.* **51**, 512–517 (2014).
71. Tlemsani, C. et al. SETD2 and DNMT3A screen in the Sotos-like syndrome French cohort. *J. Med. Genet.* **53**, 743–751 (2016).
72. Almuriekhi, M. et al. Loss-of-function mutation in APC2 causes Sotos syndrome features. *Cell Rep.* **15**, 139–134 (2015).
73. Edmunds, J. W., Mahadevan, L. C. & Clayton, A. L. Dynamic histone H3 methylation during gene induction: HYPB/Setd2 mediates all H3K36 trimethylation. *EMBO J.* **27**, 406–420 (2008).
74. Otani, J. et al. Structural basis for recognition of H3K4 methylation status by the DNA methyltransferase 3A ATRX-DNMT3-DNMT3L domain. *EMBO Rep.* **10**, 1235–1241 (2009).
75. O'Roak, B. J. et al. Multiplex targeted sequencing identifies recurrently mutated genes in autism spectrum disorders. *Science* **338**, 1619–1622 (2012).
76. Iossifov, I. et al. The contribution of de novo coding mutations to autism spectrum disorder. *Nature* **515**, 216–221 (2014).
77. Lumish, H. S., Wynn, J., Devinsky, O. & Chung, W. K. Brief report: SETD2 mutation in a child with autism, intellectual disabilities and epilepsy. *J. Autism Dev. Disord.* **45**, 3764–3770 (2015).
78. Tatton-Brown, K. et al. Mutations in the DNA methyltransferase gene DNMT3A cause an overgrowth syndrome with intellectual disability. *Nat. Genet.* **46**, 385–388 (2014).
79. Xin, B. et al. Novel DNMT3A germline mutations are associated with inherited Tatton-Brown-Rahman syndrome. *Clin. Genet.* **91**, 623–628 (2017).
80. Kosaki, R., Terashima, H., Kubota, M. & Kosaki, K. Acute myeloid leukemia-associated DNMT3A p. Arg882His mutation in a patient with tatton-Brown-Rahman overgrowth syndrome as a constitutional mutation. *Am. J. Med. Genet. A* **173**, 250–253 (2017).
81. Tatton-Brown, K. et al. The Tatton-Brown-Rahman syndrome: a clinical study of 55 individuals with de novo constitutive DNMT3A variants. *Wellcome Open Res.* **3**, 46 (2018).
82. Malan, V. et al. Distinct effects of allelic NFIX mutations on nonsense-mediated mRNA decay engender either a Sotos-like or a Marshall-Smith syndrome. *Am. J. Hum. Genet.* **87**, 189–198 (2010).
83. Klaassens, M. et al. Malan syndrome: Sotos-like overgrowth with de novo NFIX sequence variants and deletions in six new patients and a review of the literature. *Eur. J. Hum. Genet.* **23**, 610–615 (2015).
84. Martinez, F. et al. Novel mutations of NFIX gene causing Marshall-Smith syndrome or Sotos-like syndrome: one gene, two phenotypes. *Pediatr. Res.* **78**, 533–539 (2015).
85. Bateman, J. F., Boot-Handford, R. P. & Lamande, S. R. Genetic diseases of connective tissues: cellular and extracellular effects of ECM mutations. *Nat. Rev. Genet.* **10**, 173–183 (2009).
86. Mirzaa, G. et al. PIK3CA-associated developmental disorders exhibit distinct classes of mutations with variable expression and tissue distribution. *JCI Insight* **1**, e87623 (2016).
87. Tatton-Brown, K. et al. Weaver syndrome and EZH2 mutations: clarifying the clinical phenotype. *Am. J. Med. Genet. A* **161A**, 2972–2980 (2013).
88. Cao, R. et al. Role of histone H3 lysine 27 methylation in polycomb-group silencing. *Science* **298**, 1039–1043 (2002).
89. Cohen, A. S. & Gibson, W. T. EED-associated overgrowth in a second male patient. *J. Hum. Genet.* **61**, 831–834 (2016).
90. Cohen, A. S. et al. A novel mutation in EED associated with overgrowth. *J. Hum. Genet.* **60**, 339–342 (2015).
91. Cooney, E., Bi, W., Schlesinger, A. E., Vinson, S. & Potocki, L. Novel EED mutation in patient with Weaver syndrome. *Am. J. Med. Genet. A* **173A**, 541–545 (2017).
92. Neri, G., Martini-Neri, M. E., Katz, B. E. & Opitz, J. M. The Perlman syndrome: familial renal dysplasia with Wilms tumor, fetal gigantism and multiple congenital anomalies. *Am. J. Med. Genet.* **19**, 195–207 (1984).
93. Alessandri, J. L. et al. Perlman syndrome: report, prenatal findings and review. *Am. J. Med. Genet. A* **146A**, 2532–2537 (2008).
94. Astuti, D. et al. Germline mutations in DIS3L2 cause the Perlman syndrome of overgrowth and Wilms tumor susceptibility. *Nat. Genet.* **44**, 277–284 (2012).
95. Labno, A. et al. Perlman syndrome nuclease DIS3L2 controls cytoplasmic non-coding RNAs and provides surveillance pathway for maturing snRNAs. *Nucleic Acids Res.* **44**, 10437–10453 (2016).
96. Janku, F., Yap, T. A. & Meric-Bernstam, F. Targeting the PI3K pathway in cancer: are we making headway? *Nat. Rev. Clin. Oncol.* **15**, 273–291 (2018).
97. Pilarski, R. et al. Cowden syndrome and the PTEN hamartoma tumor syndrome: systematic review and revised diagnostic criteria. *J. Natl Cancer Inst.* **105**, 1607–1616 (2013).
98. Keppler-Noreuil, K. M. et al. Clinical delineation and natural history of the PIK3CA-related overgrowth spectrum. *Am. J. Med. Genet. A* **164A**, 1713–1733 (2014).
99. Biesecker, L. G. & Sapp, J. C. Proteus syndrome. *GeneReviews* https://www.ncbi.nlm.nih.gov/books/NBK99495 (updated 10 Jan 2019).
100. Mirzaa, G., Conway, R., Graham, J. M. Jr & Dobyns, W. B. PIK3CA-related segmental overgrowth. *GeneReviews* https://www.ncbi.nlm.nih.gov/books/NBK153722 (updated 15 Aug 2013).
101. Michel, M. E. et al. Causal somatic mutations in urine DNA from persons with the CLOVES subgroup of the PIK3CA-related overgrowth spectrum. *Clin. Genet.* **93**, 1075–1080 (2018).
102. Kuentz, P. et al. Molecular diagnosis of PIK3CA-related overgrowth spectrum (PROS) in 162 patients and recommendations for genetic testing. *Genet. Med.* **19**, 989–997 (2017).
103. Nathan, N., Keppler-Noreuil, K. M., Biesecker, L. G., Moss, J. & Darling, T. N. Mosaic disorders of the PI3K/PTEN/AKT/TSC/mTORC1 signaling pathway. *Dermatol. Clin.* **35**, 51–60 (2017).
104. Lindhurst, M. J. et al. A mosaic activating mutation in AKT1 associated with the Proteus syndrome. *N. Engl. J. Med.* **365**, 611–619 (2011).
105. Hussain, K. et al. An activating mutation of AKT2 and human hypoglycemia. *Science* **334**, 474 (2011).
106. Zhou, X. et al. Association of germline mutation in the PTEN tumour suppressor gene and Proteus and Proteus-like syndromes. *Lancet* **358**, 210–211 (2001).
107. Biesecker, L. G., Rosenberg, M. J., Vacha, S., Turner, J. T. & Cohen, M. M. PTEN mutations and proteus syndrome. *Lancet* **358**, 2079–2080 (2001).
108. Riviere, J. B. et al. De novo germline and postzygotic mutations in AKT3, PIK3R2 and PIK3CA cause a spectrum of related megalencephaly syndromes. *Nat. Genet.* **44**, 934–940 (2012).
109. Mirzaa, G. et al. De novo CCND2 mutations leading to stabilization of cyclin D2 cause megalencephaly-polymicrogyria-polydactyly-hydrocephalus syndrome. *Nat. Genet.* **46**, 510–515 (2014).
110. Kratz, C. P. et al. Cancer screening recommendations for individuals with Li-Fraumeni syndrome. *Clin. Cancer Res.* **23**, e38–e45 (2017).

111. Chen, S. & Parmigiani, G. Meta-analysis of BRCA1 and BRCA2 penetrance. *J. Clin. Oncol.* **25**, 1329–1333 (2007).
112. Scott, J. et al. Insulin-like growth factor-II gene expression in Wilms' tumour and embryonic tissues. *Nature* **317**, 260–262 (1985).
113. Gicquel, C. et al. Rearrangements at the 11p15 locus and overexpression of insulin-like growth factor-II gene in sporadic adrenocortical tumors. *J. Clin. Endocrinol. Metab.* **78**, 1444–1453 (1994).
114. Akmal, S. N., Yun, K., MacLay, J., Higami, Y. & Ikeda, T. Insulin-like growth factor 2 and insulin-like growth factor binding protein 2 expression in hepatoblastoma. *Hum. Pathol.* **26**, 846–851 (1995).
115. Taniguchi, T., Sullivan, M. J., Ogawa, O. & Reeve, A. E. Epigenetic changes encompassing the IGF2/H19 locus associated with relaxation of IGF2 imprinting and silencing of H19 in Wilms tumor. *Proc. Natl Acad. Sci. USA* **92**, 2159–2163 (1995).
116. Rainier, S., Dobry, C. J. & Feinberg, A. P. Loss of imprinting in hepatoblastoma. *Cancer Res.* **55**, 1836–1838 (1995).
117. Mussa, A. et al. Cancer risk in Beckwith-Wiedemann Syndrome: a systematic review and meta-analysis outlining a novel (epi)genotype specific histotype targeted screening protocol. *J. Pediatr.* **176**, 142–149 (2016).
118. Maas, S. M. et al. Phenotype, cancer risk, and surveillance in Beckwith-Wiedemann syndrome depending on molecular genetic subgroups. *Am. J. Med. Genet. A* **170A**, 2248–2260 (2016).
119. Brioude, F. et al. Revisiting Wilms tumour surveillance in Beckwith-Wiedemann syndrome with IC2 methylation loss, reply. *Eur. J. Hum. Genet.* **26**, 471–472 (2018).
120. Kalish, J. M. et al. Surveillance recommendations for children with overgrowth syndromes and predisposition to Wilms tumors and hepatoblastoma. *Clin. Cancer Res.* **23**, e115–e122 (2017).
121. Lapunzina, P. Risk of tumorigenesis in overgrowth syndromes: a comprehensive review. *Am. J. Med. Genet. C Semin. Med. Genet.* **137C**, 53–71 (2005).
122. Bennett, R. L., Swaroop, A., Troche, C. & Licht, J. D. The role of nuclear receptor-binding SET domain family histone lysine methyltransferases in cancer. *Cold Spring Harb. Perspect. Med.* **7**, a026708 (2017).
123. Nakagawa, M. & Kitabayashi, I. Oncogenic roles of enhancer of zeste homolog 1/2 in hematological malignancies. *Cancer Sci.* **109**, 2342–2348 (2018).
124. Villani, A. et al. Recommendations for cancer surveillance in individuals with RASopathies and other rare genetic conditions with increased cancer risk. *Clin. Cancer Res.* **23**, e83–e90 (2017).
125. Mester, J. & Eng, C. When overgrowth bumps into cancer: the PTEN-opathies. *Am. J. Med. Genet. C Semin. Med. Genet.* **163C**, 114–121 (2013).
126. Smith, J. R. et al. Thyroid nodules and cancer in children with PTEN hamartoma tumor syndrome. *J. Clin. Endocrinol. Metab.* **96**, 34–37 (2011).
127. Schultz, K. A. P. et al. PTEN, DICER1, FH, and their associated tumor susceptibility syndromes: clinical features, genetics, and surveillance recommendations in childhood. *Clin. Cancer Res.* **23**, e76–e82 (2017).
128. Daly, M. B. et al. NCCN guidelines insights: genetic/familial high-risk assessment: breast and ovarian, version 2.2017. *J. Natl Compr. Canc. Netw.* **15**, 9–20 (2017).
129. Gripp, K. W. et al. Nephroblastomatosis or Wilms tumor in a fourth patient with a somatic PIK3CA mutation. *Am. J. Med. Genet. A* **170A**, 2559–2569 (2016).
130. Baujat, G. et al. Clinical and molecular overlap in overgrowth syndromes. *Am. J. Med. Genet. C Semin. Med. Genet.* **137C**, 4–11 (2005).
131. Baujat, G. et al. Paradoxical NSD1 mutations in Beckwith-Wiedemann syndrome and 11p15 anomalies in Sotos syndrome. *Am. J. Hum. Genet.* **74**, 715–720 (2004).
132. Chang, F. et al. Molecular diagnosis of mosaic overgrowth syndromes using a custom-designed next-generation sequencing panel. *J. Mol. Diagn.* **19**, 613–624 (2017).

Author contributions
The authors contributed equally to all aspects of the article.

Competing interests
The authors declare no competing interests.