Clinical, radiological, and molecular diagnosis of pituitary diseases in short stature

Sarah Castets (1,3), Carine Villanueva (2,3) (co-1er auteur), Julia Vergier (1,3), Thierry Brue (3,4,2,6) Alexandru Saveanu (3,), Rachel Reynaud (1,3,5)

(1) Assistance Publique-Hôpitaux de Marseille *(*AP-HM), Hôpital Timone Enfants, Service de Pédiatrie Multidisciplinaire, Marseille, France
(2) Hospices Civils de Lyon (HCL), Hôpital Femme Mère Enfant (HFME), Service d'Endocrinologie pédiatrique, Bron, France
(3) Centre de Référence des Maladies Rares de l'Hypophyse HYPO, Marseille, France
(4) Assistance Publique-Hôpitaux de Marseille (AP-HM), Service d'Endocrinologie, Hôpital de la Conception, Marseille, France
(5) Aix Marseille Université, Institut National de la Santé et de la Recherche Médicale *(*INSERM), Marseille Medical Genetics (MMG), U 1251, Marseille, France
(6) Institut Marseille Maladies Rares (MarMaRa), Marseille, France

Adresse auteur correspondant : sarah.castets@ap-hm.fr

Abstract

Short stature in children can be caused by congenital pituitary disorders involving at least a growth hormone deficiency. Clinical and radiological evaluations of the index case and family history assessments are essential to guide genetic diagnostic testing and interpret results. The first line approach is panel testing of genes involved in pituitary development with variants known to be pathogenic in this context. This approach identifies a genetic cause in less than 10% of cases however. Whole exome and whole genome sequencing techniques may therefore provide original information but also raise new questions regarding the pathophysiological role of the identified variants. These new tools can make genetic counselling more complex. The role of clinicians in these interpretations is therefore important.

Keywords

Growth hormone deficiency, short stature, hypopituitarism, genetics, NGS, WES

1. Introduction

Congenital hypopituitarism is a rare cause of short stature, secondary to constitutional deficiencies in the synthesis or secretion of one or several anterior pituitary hormones involved in statural growth: growth hormone (GH deficiency, GHD), thyroid-stimulating hormone (TSH deficiency, TSHD), and gonadotropins (hypogonadotropic hypogonadism, HH). The pathophysiology is often analyzed in terms of altered pituitary development.

Pituitary ontogenesis begins with an invagination of the oral ectoderm of the floor of the stomodeum in contact with neuroectodermal tissue, and depends on the sequential expression of morphogenic factors (transcription and signaling factors) that promote cellular proliferation and differentiation in the five anterior pituitary cell lines in humans, and on their ongoing expression throughout life [1]. Any perturbation of this process, whether genetic or from the environment, that modifies the expression of one of these factors can lead to deficiencies in overall pituitary development (combined pituitary hormone deficiency, CPHD) or in specific cell lines (isolated hypopituitarism). Phenotypes tend to be more complex and extend beyond pituitary development when the affected factors are expressed early or are ubiquitous (complex syndromic diseases); conversely, expression deficiencies that occur later and only affect specifically pituitary factors lead to purely pituitary phenotypes. Our understanding of these mechanisms has improved significantly over the past three decades, with more than thirty genes having so far been implicated in syndromic and non-syndromic pituitary deficiencies [2] with sometimes identical phenotypic presentations.

Growth retardation due to GHD can therefore have a variety of clinical presentations: transient isolated GHD in childhood and adolescence or persisting into adulthood, GHD in combination with other pituitary deficiencies in non-syndromic presentations or in syndromic forms extending beyond disorders of pituitary development. Clinical workup is therefore essential in the diagnosis process and in providing genetic counselling, but also in the organization of care pathways.

In this article, we describe the diagnostic process in patients with short stature due to pituitary deficiency, with a particular focus on genetic testing.

2. Epidemiology

Congenital hypopituitarism is a rare disease and precise epidemiological data on this condition are lacking. In newborns, estimates based on TSHD screening data in the Netherlands put the prevalence of CPHD with TSHD at around 1/26 000 [3], while estimates of the prevalence of TSHD with or without another pituitary deficiency range from 1/160 000 to 1/31 000 and from 1/20 000 to 1/16 000,

respectively, in different countries [4,5]. Growth hormone deficiency is the main deficiency diagnosed in cases of CPHD and the most common (85.8% of patients) [6]. Its prevalence is also highly uncertain, with a historical estimate of 1/8 000 births [7] and a more recent estimate of 1/30 000 births for GHD in CPHD derived from the screening of TSHD in the Netherlands [3].

Epidemiological studies in adults put the prevalence of non-acquired hypopituitarism at 1/20 000 [8,9], higher than in children, suggesting that congenital hypopituitarism may be expressed later in life. Cohort studies with genetic analyses provide biased information on when hypopituitarism is diagnosed: in the neonatal period (24%), in childhood (28%), at puberty (32%), or in adulthood (7.2%), since pituitary hormone deficiencies continue to evolve into adulthood in some cases. Syndromic presentations and cases of CPHD tend to be diagnosed earlier, with a mean age of 4.9 years at diagnosis, versus 10.2 years for patients with non-syndromic isolated GHD (IGHD) [10].

In the past three decades, a growing number of genes involved in pituitary development have been associated with pituitary deficiencies. Currently however, a molecular cause is identified in fewer than 10% of patients with CPHD [6,11] and in 4–53% of patients with IGHD [12]. Very recently, the range of available techniques has expanded to include whole exome and whole genome sequencing (WES and WGS). These approaches provide important information but also raise new questions concerning the pathophysiological interpretation of variants and thus on how this should inform genetic counselling.

3. Role of clinical and radiological data in genetic diagnosis

The probability of identifying a genetic cause depend strongly on the familial or sporadic nature of the deficiency, whether it is isolated (IGHD) or combined (CPHD), and whether extra-pituitary malformations are present or not (syndromic or non-syndromic IGHD or CPHD).

3.1. Context

A family history of hypopituitarism and consanguinity significantly increases the chances of identifying a genetic cause. A pathogenic or probably pathogenic variant is identified in about 7% of cases of hypopituitarism, but this rate varies from 2.8% in sporadic forms to 29.7% in familial forms [6].

Growth hormone deficiency can be isolated or be part of more or less complete CPHD, involving anterior and/or posterior pituitary hormones. Testing should focus on different genetic abnormalities for isolated GHD (factors implicated in the final differentiation of somatotrophs, in the GH gene, or factors involved in regulating GH secretion) than for combined or syndromic deficiencies (factors implicated earlier in pituitary cell differentiation). The presence of extrapituitary developmental abnormalities (in syndromic forms) are indicative of more substantial genetic rearrangements or changes in early-expressed transcription factors.

3.2. Syndromic or combined forms

Combined or syndromic pituitary hormone deficiencies are caused by profound disorders of pituitary development and can be associated with other developmental abnormalities.

3.2.1 Clinical and radiological features

The analysis of a large cohort of patients with hypopituitarism as part of the GENHYPOPIT network revealed that 40% of patients with hypopituitarism had extrapituitary malformations: mostly ocular (11%, including 6% with optic nerve abnormalities, and 7% with globe abnormalities), while 4% had neurodevelopmental disorders, 3.4% had corpus callosum abnormalities, and 2.7% had hearing loss [6]. The main extrapituitary clinical signs and the corresponding genes to investigate are listed in Table 1.

Extracerebral malformations thus mainly occur in craniofacial or ectodermal ocular development. Indeed, the ectodermal placode of the adenohypophysis and the optic, olfactory and lens placodes all arise from the cranial panplacodal ectoderm [13].

Congenital pituitary hormone deficiencies are associated with a malformation in the hypothalamo–pituitary region in 50–100% of cases of CPHD and more rarely (29–84% of cases) in congenital IGHD [10,14,15]. The most commonly identified cerebral malformations are pituitary stalk interruption syndrome (PSIS) [16] and septo-optic dysplasia. The pituitary hormone deficiencies are then either multiple from the start or appear successively with no insight from genetic analysis as to the likely evolution of the condition. The malformations most commonly associated with GHD are an absent anterior pituitary, an ectopic posterior pituitary, and PSIS [17]. The presence of a cerebral malformation is indicative of a genetic cause affecting early neurological and hypothalamo–pituitary development. The main genetic causes are listed in Table 2.

Hypopituitarism can occur in a variety of syndromes such as CHARGE, Fanconi, Rieger, or Pallister–Hall syndrome. Neurodevelopmental disorders can either be hypomorphic forms of holoprosencephaly due to common genetic anomalies (*GLI2*, *SHH*, *FGF8*), or associated with genetic obesity (Prader–Willi, Bardet–Biedl, or Cohen syndrome; POMC. LEP, LEPR mutations) [18]. Hypopituitarism may then involve diabetes insipidus or severe childhood obesity.

Finally, the absence of MRI abnormalities does not necessarily invalidate the diagnosis of congenital hypopituitarism, but normal MRI findings should direct investigations toward other potential genetic causes (see below).

In summary, a thorough clinical and imaging workup should be performed alongside targeted questioning, including

- a clinical examination (dysmorphic features, ocular, ENT, hand or cardiac abnormalities, scoliosis, etc.)

- a neurodevelopmental evaluation for children
- cerebral MRI and pelvic and renal ultrasound and, depending on clinical data, spinal X-ray and/or cardiac ultrasonography
- family history investigations of post-axial polydactyly, congenital hearing loss, microphthalmia or other ocular abnormalities, anosmia or hyposmia. These malformations are not necessarily associated with hypopituitarism.

3.2.2. Genetic data

The genetic causes identified in syndromic CPHD are most often a *GLI2* mutation (in 2–10% of cases with an identified mutation) [11,19], and exceptionally *LHX4, HESX1, SOX3,* and *PROKR2* mutations [19].

GLI2 mutations are implicated in IGHD and CPHD with PSIS alone or associated with polydactyly, orofacial clefts or other hypomorphic manifestations of holoprosencephaly. Expressions of the same genotype are extremely variable (IGHD, CPHD and/or polydactyly), with incomplete penetrance (30–80%) [19]. Family health history collection should focus on cases of polydactyly that may not have been spontaneously reported. Mutations in the FGF pathway (*FGF8* and *FGFR1*) can in rare cases be associated with IGHD or CPHD with PSIS [20,21].

In cases of ocular malformations, *OTX2* mutations are a major cause of IGHD or CPHD with PSIS and ocular signs of variable severity, microphthalmia/anophthalmia [6,22]. *SOX3* mutations are also identified in some cases of IGHD or CPHD, but more rarely [11].

The involvement of *PROKR2* mutations in the pathophysiology of PSIS in CPHD has been suggested [23], with the absence of genotype-phenotype correlation implying oligogenic inheritance [24].

Table 1. Genes to investigate depending on extra-pituitary clinical signs possibly associated with hypopituitarism (non-exhaustive list)

Clinical signs possibly associated with hypopituitarism (non-exhaustive list)	**Genes of interest (non-exhaustive list)***

Ocular abnormalities	
Sensory or motor nystagmus, abnormal visual behavior in the preverbal stage, vision loss confirmed in the verbal stage	*FGF8, FGFR1, GLI2, HESX1,*
Anophthalmia, microphthalmia, coloboma	*OTX2, SOX2*
Midline anomalies	
Orofacial clefts, hypotelorism, nasal pyriform aperture stenosis	*FGFR1, FGF8, GLI2*
Dental anomalies	*SOX2, BMP4, HESX1*
Abnormal head and neck rotation, vertebral anomalies	*LHX3*
Polydactyly ± midface anomalies (midfacial hypoplasia, orofacial clefts, hypertelorism) (Culler-Jones syndrome)	*GLI2*
Hearing loss	*LHX3, SOX2, FGFR1*
Immune deficiency (DAVID syndrome)	*NFKB2*
Macroorchidism	*IGSF1*
CHARGE syndrome Coloboma with or without microphthalmia, cardiac involvement, intellectual disability, choanal atresia, external genital and ear anomalies, hearing loss, cranial nerve and central nervous system abnormalities	*CHD7*
Pallister-Hall syndrome Exocrine pancreatic insufficiency, nasal wing hypoplasia/aplasia, anorectal malformations, ± cardiopathy, liver involvement, hematological involvement	*GLI3*
Webb-Dattani syndrome Postnatal microcephaly, white matter lesions, postretinal blindness and hydronephrosis	*ARNT2*

*OMIM: *ARNT2* (OMIM # 615926), *BMP4* (OMIM # 112262), *CHD7* (OMIM # 214800), *FGF8* (OMIM # 600483), *FGFR1* (OMIM #136350), *GLI2* (OMIM # 615849), *GLI3* (OMIM # 146510),

HESX1 (OMIM # 601802), *IGSF1* (OMIM # 300888), *LHX3* (OMIM # 221750), *NFKB2* (OMIM# 615577), *OTX2* (OMIM # 600037), *SOX2* (OMIM # 184429)

Table 2. Genes to investigate depending on neuroradiological findings

MRI findings potentially associated with hypopituitarism	Genes of interest (non exhaustive list)*
Abnormalities in the hypothalamo-pituitary region	
Normal pituitary region or pituitary hypoplasia	IGHD: *GH1, GHRHR, GHSR, SOX3* CPHD: *PROP1, POU1F1, LHX3, IGSF1*
Ectopic posterior pituitary ± pituitary stalk interruption syndrome	*GLI2, HESX1, LHX4, SOX3, OTX2, FGF8, FGFR1, SOX2, PROKR2*
Uniform pituitary enlargement	*PROP1, LHX3*
Hypoplastic sella turcica	*LHX4*
Hypothalamic hamartoma	*GLI3*
Structural cerebral abnormalities	
Septo-optic or corpus callosum dysplasia	*HESX1, OTX2, SOX2, SOX3, GLI2, FGF8, PROKR2, FGFR1*
Olfactory bulb and/or olfactory tract agenesis	*CHD7, FGFR1, FGF8*
Holoprosencephaly, schizencephaly	*GLI2*
Chiari malformation	*HESX1, LHX4*
Cerebellar malformation	*HESX1*
Ocular abnormalities	
Microphthalmia, anophthalmia	*OTX2, SOX2, PAX6*

*OMIM: *CHD7* (OMIM # 214800), *FGF8* (OMIM # 600483), *FGFR1* (OMIM #136350), *GH1* (OMIM # 139250), *GHRHR* (OMIM # 139191), *GHSR* (OMIM # 601891), *GLI2* (OMIM # 615849), *GLI3* (OMIM # 146510), *HESX1* (OMIM # 601802), *LHX3* (OMIM # 221750), *LHX4* (# 262700), *OTX2* (OMIM # 600037), *PAX6* (OMIM # 607108), *POU1F1* (OMIM # 173110), *PROKR2* (OMIM # 607123), *PROP1* (OMIM # 601538), *SOX2* (OMIM # 184429), *SOX3* (OMIM # 313430)

The clinical and/or radiographic phenotype may therefore guide the genetic diagnosis toward one or several genes. Some signs are more likely than others to be associated with a genetic abnormality, namely hearing loss (*LHX3* mutation found in 20% of cases) [16], ocular abnormalities (*OTX2* mutation identified in around 10% of cases), and polydactyly (*GLI2*) [19]. The addition of pituitary

MRI findings and the association of signs increase the chances of finding a mutation [11]. However, there are no systematic genotype-phenotype correlations: none of the extrapituitary signs are specific, and clinical penetrance is often incomplete. In the GENHYPOPIT cohort for instance, none of the eight patients with hypopituitarism and abnormal neck rotation had an *LHX3* mutation [6] despite this sign being a classic feature of *LHX3* mutations. Consanguinity can modify phenotypic presentations, making diagnosis (and then genetic counselling) more complicated.

3.3. Non-syndromic forms: genetic features

3.3.1. Combined pituitary hormone deficiency

In the absence of associated malformations, the clinical characteristics associated with a higher probability of identifying a mutation are the combined nature of the deficiency, a greater patient–midparental height disparity, a lower GH peak, and a younger age at diagnosis [11].

PROP1 mutations are the most commonly identified genetic etiology in non-syndromic CPHD with considerable geographical variations: reported frequencies being highest in North Africa and Central and Eastern Europe (*PROP1* mutations identified in up to 17% of cases of CPHD) [11,16], with a founder effect for the three most common mutations. Hormone deficiencies can initially be isolated or dual and then combine progressively (GHD, TSHD, LH/FSHD, ACTHD and PRLD). *PROP1* mutations can also cause pituitary enlargement, which should not be confused with an adenoma. Recently, syndromic forms of *PROP1* mutations causing CPHD with PSIS or septo-optic dysplasia have been reported, with no clear pathophysiological link, and the roles of consanguinity and suspected digenetic inheritance to be confirmed [11].

The other known genetic causes of non-syndromic CPHD (such as *LHX3, LHX4, TBX19, GH1* mutations) are rarer [6,11], but the identification of a pathogenic *POU1F1* variant with a strong genotype-phenotype correlation is of high clinical relevance, since patients with this genetic abnormality do not develop ACTHD or gonadotropin deficiency [25,26]

3.4. Isolated growth hormone deficiency

In the absence of associated malformations, IGHD is indicative of a genetic abnormality affecting either the final differentiation of somatotropic cells, the expression of the growth hormone gene (*GH1*), or in the GH-releasing hormone receptor (GHRHR). Point mutations in *GH1* are the most frequently identified cause (in up to 5.5% of cases of IGHD, amounting to three quarters of cases in which an associated mutation is found) [11]. Transmission is autosomal recessive or autosomal dominant for dominant negative mutations. In patients with *GH1* mutations, IGHD can exceptionally evolve to CPHD [11]. The second most identified cause of IGHD is *GHRHR* mutations [11], with autosomal recessive transmission. The other reported genetic causes (*GHSR*) are rarer [27].

Genetic causes are currently rarely found in cases of partial GH deficiency appearing later than early childhood with no clinical or radiographic abnormalities other than (non-specific) pituitary hypoplasia, and with no family history of hypopituitarism [11]. However, partial GH deficiency does not exclude the possibility of finding a genetic cause (*GH1*, *PROP1*, *SOX3*) [11]. Growth hormone thresholds should therefore be used with caution and current guidelines are to use a more flexible approach based on the patient's medical history and growth rate, the presence of other pituitary hormone deficiencies, and genetic analysis [28].

4. Genetic diagnostic process

4.1. Why should genetic analysis be considered ?

Table 3. Benefits of genetic analysis in patient management

- *Support the diagnosis of GHD*
- *Estimate the expected response to GH*
- *Evaluate the risk of developing other pituitary hormone deficiencies and guide monitoring strategies*
- *Facilitate the screening and monitoring of other potentially associated conditions*
- *Offer families genetic counselling*
- *Improve knowledge of the pathophysiological mechanisms of hypopituitarism*

A diagnosis of GHD is classically confirmed by an association of clinical, biochemical and radiological features [28–30]. In the case of IGHD however, with no structural pituitary abnormalities, this set of criteria does not always provide diagnostic certainty, notably because of the imperfect sensitivity of the diagnostic tests. Molecular diagnosis is then beneficial in potentially confirming the diagnosis and in helping to organizing care pathways during transitions to adulthood [28,31].

Molecular diagnoses can also provide information on the potential development of other pituitary hormone deficiencies. In patients with *PROP1* mutations for instance, the possible sudden appearance of ACTHD, sometimes in adulthood, and the corresponding risk of death, warrants prolonged and regular follow-up [32]. Conversely, patients with a *POU1F1* mutation never develop ACTHD or gonadotropin deficiency.

For *PROP1* and *LHX3* mutations, pseudotumoral pituitary enlargement followed by spontaneous regression has been widely described, and knowledge of this avoids inappropriate neurosurgical treatment.

The identification of a mutation can be predictive of a good response to GH replacement therapy, with greater heigh gains in patients with GHD and an identified mutation than in those with idiopathic GHD [11].

Finally, identifying genetic mutations is important to provide appropriate genetic counselling to patients and their families.

4.2. Which analyses and for whom?

Genetic testing strategies are based firstly on the type of deficiency (IGHD or CPHD) and on the presence or not of a cerebral or extracerebral malformation in the index case or in the patient's family history. Analyses cannot focus on a specific gene however because of the lack of genotype-phenotype correlation. Next generation sequencing (NGS) of gene panels is therefore recommended, alongside an analysis of chromosome structure (array comparative genomic hybridization (CGH), fluorescence in situ hybridization (FISH) ± karyotype) for syndromic forms.

Table 4. Recommended genetic analyses depending on the type of pituitary hormone deficiency*

Syndromic isolated or combined pituitary hormone deficiency
- Panel of genes implicated in hypopituitarism (NGS) and - Analysis of copy number variations (array CGH, FISH ± karyotype)
Non-syndromic combined pituitary hormone deficiency
- Panel of genes implicated in hypopituitarism (NGS)
Isolated growth hormone deficiency manifesting before 5 years of age or familial form or persisting into adulthood
- Panel of genes implicated in hypopituitarism (NGS)

**syndromic conditions with or without hypopituitarism in patients' families and consanguinity should also be considered in these approaches*

4.2.1. NGS and array CGH

Next-generation sequencing identifies point mutations or copy number variations in panels of genes analyzed in parallel. The composition of the gene panel depends on the patient's phenotype, and possibly their family members'. This analysis can be completed or preceded by array CGH to detect gene duplications or deletions, particularly for patients with complex syndromic forms. Genetic testing strategies should also account for the time required for array CGH and NGS in different centers. For syndromic forms in which the primary or predominant clinical presentation is not hypopituitarism,

NGS requests should first be sent to the appropriate referral center (NGS panels for intellectual disability, holoprosencephaly, or hypothalamic obesity for instance).

In France, referral molecular biology laboratories for the genetic diagnosis of hypopituitarism have established an NGS panel of at least 15 genes for CPHD (syndromic and non-syndromic forms). These genes were chosen based on epidemiological, clinical and pathophysiological criteria, and on the presence of a high enough number of reported cases for results to be of sufficient clinical relevance to guide patient management. The analysis covers both point mutations and copy number variations, which are identified in about 1% of cases [11,33]. In cases of IGHD, genes specifically involved in the somatotropic axis (*GH1*, *GHRHR* and *GHSR*) are added to the NGS panel. The composition of these panels is regularly reevaluated based on new data on potential candidate genes [34].

4.2.2. Whole exome and whole genome sequencing

Molecular causes are only identified in a minority of cases of GHD, whether isolated of combined, which suggests that other genetic factors are still to be discovered. Since 2016, targeted or whole exome/genome studies have suggested that more than thirty genes may be involved in CPHD, sometimes associated with complex syndromes [35,36].

In the absence of an identified mutation in the NGS panel, secondary analyses can be performed by sequencing the whole exome (the protein-coding region of genes) or the entire genome (i.e. including non-coding regulatory regions) by trio analysis (the index case and two first-degree relatives). This technique has been available in France since 2020 as part of the *France Médecine Génomique 2025* program for cases of hypopituitarism satisfying the PFMG2025 criteria and approved by dedicated multidisciplinary committees.

These WES and WGS data should reveal new candidate genes. Those that are clinically significant (GAD) must then be separated from those of uncertain significance (GUS) [37]. Clinically significant genes can be included in diagnostic reasoning and genetic counselling, whereas GUS are still the subject of more or less advanced studies and variants cannot be described as pathogenic [37].

To interpret rare variants of genes implicated in hypopituitarism, bioinformatic analyses should be supplemented with an analysis of clinical and epidemiological data, and possibly also additional functional studies, to reach a meaningful interpretation: phenotype-genotype correlations and familial segregation are essential to interpret their pathogenicity. In some cases, analyses are inconclusive as to the implication of a variant allele in the pituitary phenotype [16]; this variant is then considered of uncertain significance and should not immediately be considered in genetic counselling. These results can be reviewed however, and genetic counselling adapted, in light of an accumulation of data and collaborations with growth and/or genetic specialists. Accumulations of whole genome analyses are also an opportunity to increase knowledge of the mechanisms involved in hypopituitarism, through the discovery of new causal genes, or of pathogenic variants in genes or regulatory regions.

5. Conclusion

Short stature in children can be caused by pituitary pathologies involving at least one form of growth hormone deficiency. Etiological investigations should identify whether this is part of a syndromic presentation, in the index case or their family. Genetic investigations based on this phenotypic analysis can then proceed thorough the sequencing of panels of genes, albeit with low hit rates. The use of whole exome and whole genome sequencing promises new progress in the understanding of the mechanisms involved in pituitary pathologies. However, the roles of variants identified with these approaches are difficult to interpret because of the variable and incomplete penetrance of the genes involved, the probable oligogenic and epigenetic nature of the mechanisms, and low levels of genotype-phenotype correlation. Genetic counselling is therefore complicated. Nevertheless, beyond improvements in pathophysiological understanding and despite low genotype-phenotype correlations, genetic diagnosis is helpful for clinicians. It contributes to confirming diagnoses of GHD and can provide information on how the deficiency will evolve into adulthood and whether prolonged replacement therapy will be required. In some cases also, genetic testing provides crucial information on the possible appearance of other pituitary hormone deficiencies or associated conditions and helps guide long-term monitoring strategies.

References

[1] S.W. Davis, B.S. Ellsworth, M.I. Peréz Millan, P. Gergics, V. Schade, N. Foyouzi, M.L. Brinkmeier, A.H. Mortensen, S.A. Camper, Pituitary gland development and disease: from stem cell to hormone production, Curr. Top. Dev. Biol. 106 (2013) 1–47. https://doi.org/10.1016/B978-0-12-416021-7.00001-8.

[2] Q. Fang, A.S. George, M.L. Brinkmeier, A.H. Mortensen, P. Gergics, L.Y.M. Cheung, A.Z. Daly, A. Ajmal, M.I. Pérez Millán, A.B. Ozel, J.O. Kitzman, R.E. Mills, J.Z. Li, S.A. Camper, Genetics of Combined Pituitary Hormone Deficiency: Roadmap into the Genome Era, Endocr. Rev. 37 (2016) 636–675. https://doi.org/10.1210/er.2016-1101.

[3] D.A. van Tijn, J.J.M. de Vijlder, B. Verbeeten, P.H. Verkerk, T. Vulsma, Neonatal detection of congenital hypothyroidism of central origin, J. Clin. Endocrinol. Metab. 90 (2005) 3350–3359. https://doi.org/10.1210/jc.2004-2444.

[4] C. Peters, A.S.P. van Trotsenburg, N. Schoenmakers, DIAGNOSIS OF ENDOCRINE DISEASE: Congenital hypothyroidism: update and perspectives, Eur. J. Endocrinol. 179 (2018) R297–R317. https://doi.org/10.1530/EJE-18-0383.

[5] P. van Trotsenburg, A. Stoupa, J. Léger, T. Rohrer, C. Peters, L. Fugazzola, A. Cassio, C. Heinrichs, V. Beauloye, J. Pohlenz, P. Rodien, R. Coutant, G. Szinnai, P. Murray, B. Bartés, D. Luton, M. Salerno, L. de Sanctis, M. Vigone, H. Krude, L. Persani, M. Polak, Congenital Hypothyroidism: A 2020-2021 Consensus Guidelines Update-An ENDO-European Reference Network Initiative Endorsed by the European Society for Pediatric Endocrinology and the

European Society for Endocrinology, Thyroid Off. J. Am. Thyroid Assoc. 31 (2021) 387–419. https://doi.org/10.1089/thy.2020.0333.
[6] N. Jullien, A. Saveanu, J. Vergier, E. Marquant, M.H. Quentien, F. Castinetti, N. Galon-Faure, R. Brauner, Z. Marrakchi Turki, M. Tauber, M. El Kholy, A. Linglart, P. Rodien, N.S. Fedala, I. Bergada, C. Cortet-Rudelli, M. Polak, M. Nicolino, C. Stuckens, A. Barlier, T. Brue, R. Reynaud, Genhypopit Network, Clinical lessons learned in constitutional hypopituitarism from two decades of experience in a large international cohort, Clin. Endocrinol. (Oxf.). 94 (2021) 277–289. https://doi.org/10.1111/cen.14355.
[7] J.A. Phillips, J.D. Cogan, Genetic basis of endocrine disease. 6. Molecular basis of familial human growth hormone deficiency, J. Clin. Endocrinol. Metab. 78 (1994) 11–16. https://doi.org/10.1210/jcem.78.1.8288694.
[8] M. Regal, C. Páramo, S.M. Sierra, R.V. Garcia-Mayor, Prevalence and incidence of hypopituitarism in an adult Caucasian population in northwestern Spain, Clin. Endocrinol. (Oxf.). 55 (2001) 735–740. https://doi.org/10.1046/j.1365-2265.2001.01406.x.
[9] T. Rosén, B.A. Bengtsson, Premature mortality due to cardiovascular disease in hypopituitarism, Lancet Lond. Engl. 336 (1990) 285–288. https://doi.org/10.1016/0140-6736(90)91812-o.
[10] C. Deal, C. Hasselmann, R.W. Pfäffle, A.G. Zimmermann, C.A. Quigley, C.J. Child, E.P. Shavrikova, G.B. Cutler, W.F. Blum, Associations between pituitary imaging abnormalities and clinical and biochemical phenotypes in children with congenital growth hormone deficiency: data from an international observational study, Horm. Res. Paediatr. 79 (2013) 283–292. https://doi.org/10.1159/000350829.
[11] W.F. Blum, J. Klammt, S. Amselem, H.M. Pfäffle, M. Legendre, M.-L. Sobrier, M.-P. Luton, C.J. Child, C. Jones, A.G. Zimmermann, C.A. Quigley, G.B. Cutler, C.L. Deal, J. Lebl, R.G. Rosenfeld, J.S. Parks, R.W. Pfäffle, Screening a large pediatric cohort with GH deficiency for mutations in genes regulating pituitary development and GH secretion: Frequencies, phenotypes and growth outcomes, EBioMedicine. 36 (2018) 390–400. https://doi.org/10.1016/j.ebiom.2018.09.026.
[12] C. Yu, B. Xie, Z. Zhao, S. Zhao, L. Liu, X. Cheng, X. Li, B. Cao, J. Shao, J. Chen, H. Zhao, Z. Yan, C. Su, Y. Niu, Y. Song, L. Wei, Y. Wang, X. Ren, L. Fan, B. Zhang, C. Li, B. Gui, Y. Zhang, L. Wang, S. Chen, J. Zhang, Z. Wu, C. Gong, X. Fan, N. Wu, Whole Exome Sequencing Uncovered the Genetic Architecture of Growth Hormone Deficiency Patients, Front. Endocrinol. 12 (2021) 711991. https://doi.org/10.3389/fendo.2021.711991.
[13] G. Schlosser, Induction and specification of cranial placodes, Dev. Biol. 294 (2006) 303–351. https://doi.org/10.1016/j.ydbio.2006.03.009.
[14] V. Pampanini, S. Pedicelli, J. Gubinelli, G. Scirè, M. Cappa, B. Boscherini, S. Cianfarani, Brain Magnetic Resonance Imaging as First-Line Investigation for Growth Hormone Deficiency Diagnosis in Early Childhood, Horm. Res. Paediatr. 84 (2015) 323–330. https://doi.org/10.1159/000439590.
[15] L. van Iersel, H.M. van Santen, G.R.J. Zandwijken, N. Zwaveling-Soonawala, A.C.S. Hokken-Koelega, A.S.P. van Trotsenburg, Low FT4 Concentrations around the Start of Recombinant Human Growth Hormone Treatment: Predictor of Congenital Structural Hypothalamic-Pituitary Abnormalities?, Horm. Res. Paediatr. 89 (2018) 98–107. https://doi.org/10.1159/000486033.
[16] T. Brue, A. Saveanu, N. Jullien, T. Fauquier, F. Castinetti, A. Enjalbert, A. Barlier, R. Reynaud, Lessons from monogenic causes of growth hormone deficiency, Ann. Endocrinol. 78 (2017) 77–79. https://doi.org/10.1016/j.ando.2017.04.001.
[17] M.A. Kalina, B. Kalina-Faska, K. Gruszczyńska, J. Baron, E. Małecka-Tendera, Usefulness of magnetic resonance findings of the hypothalamic-pituitary region in the management of short children with growth hormone deficiency: evidence from a longitudinal study, Childs Nerv. Syst. ChNS Off. J. Int. Soc. Pediatr. Neurosurg. 28 (2012) 121–127. https://doi.org/10.1007/s00381-011-1594-7.
[18] S. Courbage, C. Poitou, J. Le Beyec-Le Bihan, A. Karsenty, J. Lemale, V. Pelloux, J.-M. Lacorte, J.-C. Carel, N. Lecomte, C. Storey, G. De Filippo, M. Coupaye, J.-M. Oppert, P. Tounian, K. Clément, B. Dubern, Implication of Heterozygous Variants in Genes of the Leptin-Melanocortin Pathway in Severe Obesity, J. Clin. Endocrinol. Metab. 106 (2021) 2991–3006. https://doi.org/10.1210/clinem/dgab404.

[19] K.A. Bear, B.D. Solomon, S. Antonini, I.J.P. Arnhold, M.M. França, E.H. Gerkes, D.K. Grange, D.W. Hadley, J. Jääskeläinen, S.S. Paulo, P. Rump, C.A. Stratakis, E.M. Thompson, M. Willis, T.L. Winder, A.A.L. Jorge, E. Roessler, M. Muenke, Pathogenic mutations in GLI2 cause a specific phenotype that is distinct from holoprosencephaly, J. Med. Genet. 51 (2014) 413–418. https://doi.org/10.1136/jmedgenet-2013-102249.

[20] T. Raivio, M. Avbelj, M.J. McCabe, C.J. Romero, A.A. Dwyer, J. Tommiska, G.P. Sykiotis, L.C. Gregory, D. Diaczok, V. Tziaferi, M.W. Elting, R. Padidela, L. Plummer, C. Martin, B. Feng, C. Zhang, Q.-Y. Zhou, H. Chen, M. Mohammadi, R. Quinton, Y. Sidis, S. Radovick, M.T. Dattani, N. Pitteloud, Genetic Overlap in Kallmann Syndrome, Combined Pituitary Hormone Deficiency, and Septo-Optic Dysplasia, J. Clin. Endocrinol. Metab. 97 (2012) E694–E699. https://doi.org/10.1210/jc.2011-2938.

[21] İ.M. Erbaş, A. Paketçi, S. Acar, L.D. Kotan, K. Demir, A. Abacı, E. Böber, A nonsense variant in FGFR1: a rare cause of combined pituitary hormone deficiency, J. Pediatr. Endocrinol. Metab. JPEM. 33 (2020) 1613–1615. https://doi.org/10.1515/jpem-2020-0029.

[22] L.C. Gregory, P. Gergics, M. Nakaguma, H. Bando, G. Patti, M.J. McCabe, Q. Fang, Q. Ma, A.B. Ozel, J.Z. Li, M.M. Poina, A.A.L. Jorge, A.F.F. Benedetti, A.M. Lerario, I.J.P. Arnhold, B.B. Mendonca, M. Maghnie, S.A. Camper, L.R.S. Carvalho, M.T. Dattani, The phenotypic spectrum associated with OTX2 mutations in humans, Eur. J. Endocrinol. 185 (2021) 121–135. https://doi.org/10.1530/EJE-20-1453.

[23] R. Reynaud, S.A. Jayakody, C. Monnier, A. Saveanu, J. Bouligand, A.-M. Guedj, G. Simonin, P. Lecomte, A. Barlier, P. Rondard, J.P. Martinez-Barbera, A. Guiochon-Mantel, T. Brue, PROKR2 Variants in Multiple Hypopituitarism with Pituitary Stalk Interruption, J. Clin. Endocrinol. Metab. 97 (2012) E1068–E1073. https://doi.org/10.1210/jc.2011-3056.

[24] S.E. McCormack, D. Li, Y.J. Kim, J.Y. Lee, S.-H. Kim, R. Rapaport, M.A. Levine, Digenic Inheritance of PROKR2 and WDR11 Mutations in Pituitary Stalk Interruption Syndrome, J. Clin. Endocrinol. Metab. 102 (2017) 2501–2507. https://doi.org/10.1210/jc.2017-00332.

[25] A.P. Otto, M.M. França, F.A. Correa, E.F. Costalonga, C.C. Leite, B.B. Mendonca, I.J.P. Arnhold, L.R.S. Carvalho, A.A.L. Jorge, Frequent development of combined pituitary hormone deficiency in patients initially diagnosed as isolated growth hormone deficiency: a long term follow-up of patients from a single center, Pituitary. 18 (2015) 561–567. https://doi.org/10.1007/s11102-014-0610-9.

[26] G. Binder, D. Schnabel, T. Reinehr, R. Pfäffle, H.-G. Dörr, M. Bettendorf, B. Hauffa, J. Woelfle, Evolving pituitary hormone deficits in primarily isolated GHD: a review and experts' consensus, Mol. Cell. Pediatr. 7 (2020) 16. https://doi.org/10.1186/s40348-020-00108-2.

[27] O. Hess, O. Admoni, M. Khayat, G. Elias, T. Almagor, S.-A. Shalev, Y. Tenenbaum-Rakover, Ghrelin and growth hormone secretagogue receptor (GHSR) genes are not commonly involved in growth or weight abnormalities in an Israeli pediatric population, J. Pediatr. Endocrinol. Metab. JPEM. 25 (2012) 537–540. https://doi.org/10.1515/jpem-2012-0044.

[28] A. Grimberg, S.A. DiVall, C. Polychronakos, D.B. Allen, L.E. Cohen, J.B. Quintos, W.C. Rossi, C. Feudtner, M.H. Murad, Drug and Therapeutics Committee and Ethics Committee of the Pediatric Endocrine Society, Guidelines for Growth Hormone and Insulin-Like Growth Factor-I Treatment in Children and Adolescents: Growth Hormone Deficiency, Idiopathic Short Stature, and Primary Insulin-Like Growth Factor-I Deficiency, Horm. Res. Paediatr. 86 (2016) 361–397. https://doi.org/10.1159/000452150.

[29] P.F. Collett-Solberg, G. Ambler, P.F. Backeljauw, M. Bidlingmaier, B.M.K. Biller, M.C.S. Boguszewski, P.T. Cheung, C.S.Y. Choong, L.E. Cohen, P. Cohen, A. Dauber, C.L. Deal, C. Gong, Y. Hasegawa, A.R. Hoffman, P.L. Hofman, R. Horikawa, A.A.L. Jorge, A. Juul, P. Kamenický, V. Khadilkar, J.J. Kopchick, B. Kriström, M. de L.A. Lopes, X. Luo, B.S. Miller, M. Misra, I. Netchine, S. Radovick, M.B. Ranke, A.D. Rogol, R.G. Rosenfeld, P. Saenger, J.M. Wit, J. Woelfle, Diagnosis, Genetics, and Therapy of Short Stature in Children: A Growth Hormone Research Society International Perspective, Horm. Res. Paediatr. 92 (2019) 1–14. https://doi.org/10.1159/000502231.

[30] Déficit hypophysaire congénital, Haute Aut. Santé. (n.d.). https://www.has-sante.fr/jcms/p_3301031/fr/deficit-hypophysaire-congenital (accessed December 6, 2021).

[31] P.E. Clayton, R.C. Cuneo, A. Juul, J.P. Monson, S.M. Shalet, M. Tauber, European Society of Paediatric Endocrinology, Consensus statement on the management of the GH-treated adolescent in the transition to adult care, Eur. J. Endocrinol. 152 (2005) 165–170. https://doi.org/10.1530/eje.1.01829.

[32] S. Pekic, M. Doknic, D. Miljic, A. Saveanu, R. Reynaud, A. Barlier, T. Brue, V. Popovic, Case seminar: a young female with acute hyponatremia and a sellar mass, Endocrine. 40 (2011) 325–331. https://doi.org/10.1007/s12020-011-9516-8.

[33] E. Bertko, J. Klammt, P. Dusatkova, M. Bahceci, N. Gonc, L. Ten Have, N. Kandemir, G. Mansmann, B. Obermannova, W. Oostdijk, H. Pfäffle, D. Rockstroh-Lippold, M. Schlicke, A.K. Tuzcu, R. Pfäffle, Combined pituitary hormone deficiency due to gross deletions in the POU1F1 (PIT-1) and PROP1 genes, J. Hum. Genet. 62 (2017) 755–762. https://doi.org/10.1038/jhg.2017.34.

[34] S. Richards, N. Aziz, S. Bale, D. Bick, S. Das, J. Gastier-Foster, W.W. Grody, M. Hegde, E. Lyon, E. Spector, K. Voelkerding, H.L. Rehm, ACMG Laboratory Quality Assurance Committee, Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology, Genet. Med. Off. J. Am. Coll. Med. Genet. 17 (2015) 405–424. https://doi.org/10.1038/gim.2015.30.

[35] R. Brauner, J. Bignon-Topalovic, A. Bashamboo, K. McElreavey, Pituitary stalk interruption syndrome is characterized by genetic heterogeneity, PloS One. 15 (2020) e0242358. https://doi.org/10.1371/journal.pone.0242358.

[36] X. Fang, Y. Zhang, J. Cai, T. Lu, J. Hu, F. Yuan, P. Chen, Identification of novel candidate pathogenic genes in pituitary stalk interruption syndrome by whole-exome sequencing, J. Cell. Mol. Med. 24 (2020) 11703–11717. https://doi.org/10.1111/jcmm.15781.

[37] L.J.H. Bean, B. Funke, C.M. Carlston, J.L. Gannon, S. Kantarci, B.L. Krock, S. Zhang, P. Bayrak-Toydemir, ACMG Laboratory Quality AssuranceCommittee, Diagnostic gene sequencing panels: from design to report-a technical standard of the American College of Medical Genetics and Genomics (ACMG), Genet. Med. Off. J. Am. Coll. Med. Genet. 22 (2020) 453–461. https://doi.org/10.1038/s41436-019-0666-z.