

UNIVERSITY OF BIRMINGHAM

Research at Birmingham

Congenital Adrenal Hyperplasia: The Lancet Seminar Series

EI-Maouche, Diala; Arlt, Wiebke; Merke, Deborah P

DOI:

[10.1016/S0140-6736\(17\)31431-9](https://doi.org/10.1016/S0140-6736(17)31431-9)

License:

Creative Commons: Attribution-NonCommercial-NoDerivs (CC BY-NC-ND)

Document Version

Peer reviewed version

Citation for published version (Harvard):

EI-Maouche, D, Arlt, W & Merke, DP 2017, 'Congenital Adrenal Hyperplasia: The Lancet Seminar Series', The Lancet. [https://doi.org/10.1016/S0140-6736\(17\)31431-9](https://doi.org/10.1016/S0140-6736(17)31431-9)

[Link to publication on Research at Birmingham portal](#)

Publisher Rights Statement:

Checked 1/6/2017

General rights

Unless a licence is specified above, all rights (including copyright and moral rights) in this document are retained by the authors and/or the copyright holders. The express permission of the copyright holder must be obtained for any use of this material other than for purposes permitted by law.

- Users may freely distribute the URL that is used to identify this publication.
- Users may download and/or print one copy of the publication from the University of Birmingham research portal for the purpose of private study or non-commercial research.
- User may use extracts from the document in line with the concept of 'fair dealing' under the Copyright, Designs and Patents Act 1988 (?)
- Users may not further distribute the material nor use it for the purposes of commercial gain.

Where a licence is displayed above, please note the terms and conditions of the licence govern your use of this document.

When citing, please reference the published version.

Take down policy

While the University of Birmingham exercises care and attention in making items available there are rare occasions when an item has been uploaded in error or has been deemed to be commercially or otherwise sensitive.

If you believe that this is the case for this document, please contact UBIRA@lists.bham.ac.uk providing details and we will remove access to the work immediately and investigate.

Congenital Adrenal Hyperplasia: The Lancet

Seminar Series

Diala El-Maouche¹, MD, MS, Wiebke Arlt², MD, DSc, FRCP, Deborah P.

Merke^{1,3}, MD, MS

¹National Institutes of Health Clinical Center, Bethesda, Maryland, USA 20892;

²Institute of Metabolism and Systems Research (IMSR), University of Birmingham & Centre for Endocrinology, Diabetes and Metabolism (CEDAM), Birmingham Health Partners, Birmingham, UK; ³The *Eunice Kennedy Shriver* National Institute of Child Health and Human Development, Bethesda, Maryland, USA 20892.

Corresponding author:

Dr. Deborah P. Merke

10 Center Drive, Room 1-2740

Bethesda, MD 20892-1932

Email: DMerke@nih.gov

Search Strategy and Selection Criteria:

We searched the Cochrane Library (January 2010-September 2016), MEDLINE (January 2010-September 2016), and EMBASE (January 2010-September 2016). Keywords and controlled vocabulary and their synonyms were used when appropriate. We used variations of the search term “congenital adrenal hyperplasia” in combination with the terms “diagnosis/diagnostics”, “genetics”, “genomics”, “adrenal crisis”, “glucocorticoid”, “mineralocorticoid”, “gene therapy”, “quality of life”, “well-being”, “screening”, “metabolomics”, “prenatal”, “antenatal”, “bone mineral density”, “tumor”, “pregnancy”, “treatment/therapy/ therapeutic”, “fertility/fecundity”, “surgery”, “management”, “metabolic”, “complications”. We largely selected publications in the past 5 years, but did not exclude commonly referenced and highly regarded older publications. We also searched the reference lists of articles identified by this search strategy and selected those we judged relevant. Review articles and book chapters are cited to provide readers with more details and more references than this Seminar has room for.

Introduction

In 1865, Luigi De Crecchio, an Italian pathologist, described the life of a male, whose autopsy revealed female internal anatomy and large adrenal glands, representing the first known case of presumed congenital adrenal hyperplasia (CAH)¹ (Figure 1). However, treatment for CAH was not introduced until almost a century later when cortisone was given for what was then known as the adrenogenital syndrome.¹⁷⁻¹⁹

CAH comprises a group of seven autosomal recessive diseases caused by a defect in any one of the several steps leading to cortisol biosynthesis: 21-hydroxylase (21OH), 11 β -hydroxylase (11 β OH), 17 α -hydroxylase/ 17,20-lyase (17OH), 3 β -hydroxysteroid dehydrogenase type 2 (3 β HSD2), steroidogenic acute regulatory protein (StAR), P450 cholesterol side-chain cleavage enzyme (SCC), and P450 Oxidoreductase (POR). The pathophysiology and genetics of the various types of CAH have been elucidated in the past 50 years. Multiple hormonal imbalances occur and CAH manifests with a range of severity clinically and biochemically, with or without alterations in glucocorticoid, mineralocorticoid, and sex steroid production. Both severe (classic) and mild (nonclassic) forms have been described.

Over 95% of CAH is due to 21OH deficiency,²⁰ characterized by impaired cortisol and aldosterone production and androgen excess. Life-saving neonatal screening for classic CAH due to 21OH deficiency was first performed in Alaska in 1977⁷ and is currently being carried out worldwide in over 40 countries, including all 50 US states since 2009, though not yet implemented in the UK.^{7,20-22} Although all types of classic CAH are rare orphan diseases, the nonclassic form of 21OH deficiency is estimated to be one of the most common autosomal recessive disorders.²³

Despite these advances, CAH remains one of the most challenging endocrine disorders to diagnose, manage and treat because of the direct and indirect effects on steroidogenic pathways and the rarity of these conditions. Advances in genetics, metabolomics and treatment strategies continue to improve our understanding of these complex diseases and aim to improve patient outcome. Following is an overview of these complex disorders.

Genetics and Pathophysiology of the Various Forms of CAH

All types of CAH are monogenetic and autosomal recessive. Most patients are compound heterozygotes, meaning they have different mutations in the two alleles. The clinical manifestation follows the less severely mutated allele, and, in general, there is good genotype-phenotype correlation.^{24,25}

Adrenal steroidogenesis occurs by a series of steps facilitated by the zone specific enzyme expression and different types of CAH interrupt this process at distinct branchpoints. In addition to the classical well-established steroidogenesis pathway, an alternative pathway to active androgen biosynthesis exists (termed “backdoor” pathway),^{26,27} which may play a role in the pathophysiology of CAH (Figure 2A). The clinical manifestation of CAH is closely related to the type and severity of the impairment.

21-hydroxylase (21OH) deficiency

The gene for 21OH, *CYP21A2*, is located within the human leukocyte antigen class III region of chromosome 6 (Table 1). *CYP21A2* and a homologous pseudogene, *CYP21A1P*, lie approximately 30 kb apart. Meiotic recombination events are common in this genomic region

because of the high degree of sequence homology between duplicated genes. Approximately 95% of *CYP21A2* disease causing mutations are *CYP21A1P* derived variants or deletions due to recombination events.^{24,35}

Defective 21-hydroxylation results in elevated precursors, most notably 17-hydroxyprogesterone (17OHP), which is used for diagnosis, and decreased glucocorticoid and mineralocorticoid synthesis (Figure 2B). ACTH-stimulated androgen production occurs because there is no block in the pathway leading to adrenal androgens.

Conventionally, classic 21OH deficiency is subclassified into salt-wasting (SW) and simple virilizing (SV) forms, reflecting the severity of aldosterone deficiency. Mutations which completely inactivate *CYP21A2* result in the SW phenotype, which, without neonatal screening, presents in the first 2 weeks of life with a life-threatening adrenal crisis (Table 2).²⁰ Patients with classic SV CAH have mutations that retain 1-2 % of 21OH activity, and minimal aldosterone production prevents a neonatal crisis. Excess fetal adrenal androgen exposure results in virilization of the external genitalia of 46,XX classic patients (SW and SV) (Figure 3A). Without neonatal screening, SV males are diagnosed as toddlers with signs and symptoms of androgen excess. Postnatal excess androgen leads to premature pubic hair and rapid skeletal growth. Patients with the nonclassic form retain up to 50 percent of enzyme activity and mostly do not have adrenal insufficiency, but may have partial glucocorticoid deficiency, and female genitalia are normal. Patients may present with mild androgen excess or have little or no symptoms. In fact, the term cryptic CAH was coined to define patients with nonclassic CAH who are identified by family genetic studies, but are otherwise asymptomatic.³⁶

11-hydroxylase (11 β OH) deficiency

CAH due to 11 β OH deficiency is due to *CYP11B1* mutations (Table 1). *CYP11B1* functions in the zona fasciculata to convert 11-deoxycortisol to cortisol and deoxycorticosterone (DOC) to corticosterone, under ACTH regulation (Figure 2C). The majority of *CYP11B1* mutations correspond to minimal or absent enzyme activity resulting in a classic phenotype.³⁷

Impaired 11-hydroxylation results in decreased corticosterone and cortisol synthesis, with subsequent increase in ACTH and excess androgens, because of shunting of the pathway towards androgen production. Normally corticosterone and DOC production by *CYP11B1* transcription in the adrenal zona fasciculata is minimal, but DOC levels can rise dramatically under the influence of ACTH.³⁸ DOC is a weak mineralocorticoid, however elevated DOC suppresses the renin-angiotensin system and results in extracellular fluid volume expansion, hypertension, low plasma renin activity and low aldosterone levels, although the ability to produce aldosterone remains. These effects may not occur in the neonatal period due to the renal mineralocorticoid resistance that is present in the first few months of life. Clinically, patients present similar to 21OH deficiency, with signs of androgen excess, but also have hypertension rather than salt loss (Table 2).²⁸ A nonclassic form of *CYP11B1* deficiency exists but is very rare.³⁹

17 α -hydroxylase/ 17,20-lyase (17OH) deficiency

The *CYP17A1* gene encodes an enzyme that expresses both 17 α -OH and 17,20-lyase activities (Table 1). Due to its location in the steroidogenic pathway, severe mutations impair adrenal and gonadal sex steroid production (Figure 2D), which causes sexual infantilism and pubertal failure

(Table 2).³¹ There is a block in DHEA production, which prevents adrenarche and the development of pubic and axillary hair. *CYP17A1* is expressed in the zona fasciculata and zona reticularis, not in the zona glomerulosa. Therefore, ACTH-mediated steroidogenesis results in elevated DOC and corticosterone. High levels of DOC cause sodium retention, hypertension and hypokalemia, with suppression of aldosterone production. Corticosterone, which has glucocorticoid activity, prevents adrenal crisis, although cortisol production is low or absent. Both 46,XX and 46,XY patients have female external genitalia, and usually present during puberty as females with lack of secondary sexual characteristics, hypergonadotropic hypogonadism and low-renin hypertension (Table 2). Isolated 17,20-lyase- deficiency has been reported⁴⁰ but is extremely rare and in truly isolated form is caused by mutations in cytochrome b5 (*CYB5A*), the co-factor needed by *CYP17A1* to exert 17,20 lyase activity.⁴¹ Although phenotype variability occurs, a nonclassic form with subtle clinical manifestations has not been defined.

3 β -hydroxysteroid dehydrogenase type 2 (3 β HSD2) deficiency

3 β -hydroxysteroid dehydrogenase exists in two isoforms, type 1 (3 β HSD1) and type 2 (3 β HSD2), encoded by *HSD3B1* and *HSD3B2* respectively (Table 1). *HSD3B2* is highly expressed in the adrenals and gonads, while *HSD3B1* is expressed in the placenta and peripheral tissues.

Impaired 3 β HSD2 results in decreased aldosterone, cortisol, and androstenedione, with a subsequent increase in renin, ACTH, and DHEA (Figure 2E). DHEA can be converted to testosterone by extraadrenal 3 β HSD1. Patients present in infancy with a salt-wasting adrenal crisis, underdeveloped 46,XY genitalia and rarely 46,XX virilization (Table 2).

The hormonal criteria for diagnosing 3 β HSD2 deficiency has changed over the past three decades because the initial studies identifying a possible nonclassic form were not based on genetic findings and subsequent genetic studies failed to confirm the diagnosis.⁴²⁻⁴⁴ Nonclassic 3 β HSD2 deficiency exists, but is extremely rare.

P450 Oxidoreductase (POR) deficiency

POR plays a key role in the electron transport that occurs in the endoplasmic reticulum and several enzymes, including 17OH, 21OHD, and aromatase, depend on POR for their catalytic activity (Figure 2F). The discovery of POR deficiency in 2004^{15,16} provided an explanation for multiple hormonal deficiency initially known as apparent combined 17OH and 21OH deficiency. Lack of placental aromatization of fetal androgens could contribute to the virilization observed in some mothers carrying affected babies. However, the production of androgens via an alternative pathway to the most potent androgen, non-aromatizable 5 α -dihydrotestosterone (DHT), may also explain the prenatal virilization of affected females, while postnatally affected individuals invariably display sex steroid deficiency.¹⁶ POR also acts also as an electron donor to cytochrome P450 (CYP) enzymes other than steroidogenic CYP enzymes, explaining POR deficiency-associated changes in drug metabolism⁴⁵ and the pathogenesis of the often observed skeletal dysplasia.⁴⁶

Most *POR* mutations retain some enzymatic function; homozygous mutations with complete loss of function may not be viable, as seen in rodent models.⁴⁷ Presentation varies from mildly affected adults with amenorrhea and polycystic ovaries (females) or androgen deficiency (males), to severe hormone disturbances causing atypical genitalia in both 46,XX and 46,XY

(Table 1, 2). 46,XX virilization does not progress, and patients postnatally have sex steroid deficiency. Craniosynostosis, radioulnar or radiohumeral synostosis, midface hypoplasia, and other skeletal manifestations resembling the Antley-Bixler syndrome may occur.

Generally, POR deficient patients do not have mineralocorticoid deficiency as impairment of 17-hydroxylase increases mineralocorticoid intermediates, and affected adults may develop hypertension. Patients have variable cortisol response to cosyntropin testing, with most patients requiring either permanent or stress dose glucocorticoid coverage.³²

Lipoid CAH

Classic lipoid CAH is characterized by deficiency of all steroid hormones and is due to *StAR* mutations (Table 1, Figure 2G). *StAR* regulates the transfer of cholesterol from the outer to the inner mitochondrial membrane, a key step in steroidogenesis initiation. When cholesterol cannot be mobilized, adrenal lipid droplets accumulate and are seen on autopsy, thus the name lipoid CAH. Lipoid CAH is one of rarest forms of CAH, resulting in neonatal crisis, and both 46,XX and 46,XY infants have female external genitalia (Table 2).⁴⁸ Later presentation up to one year of age has been described.⁴⁹

The pathogenesis of lipoid CAH is explained by a two-hit model: first, loss of *StAR* leads to the accumulation of intracellular cholesterol and cholesterol esters, and the second hit arises from destruction of the cellular function by the accumulated products.^{48,50,51} This mechanism explains some unusual phenotype. Spontaneous puberty has been described in 46,XX patients, because of minimal ovarian *StAR* expression.⁴⁸ The ovaries are quiescent during fetal life and childhood and therefore toxic accumulation of cholesterol can be delayed until adolescence.

A nonclassic form was first described in 2006⁵² associated with mutations that retain approximately 20-30% of StAR activity. Most of these cases were initially misdiagnosed as Addison's disease or isolated familial glucocorticoid deficiency.⁵³ Nonclassic patients may present as toddlers or as late as adulthood, with insidious onset of glucocorticoid deficiency, hyperpigmentation and high ACTH (but mostly intact mineralocorticoid function). A wide variation in gonadal function has been reported ranging from hypergonadotropic hypogonadism to normal.⁴⁸ Similarly, 46,XY nonclassic patients may have normal male genitalia and normal puberty or be born with atypical genitalia.⁵⁴

Cholesterol side-chain cleavage enzyme (SCC) deficiency

P450 cholesterol side chain cleavage enzyme (SCC) is the first and rate-limiting step in the steroidogenic pathway (Figure 2G), encoded by *CYP11A1* (Table 1) and is clinically and biochemically identical to lipoid CAH (Table 2); however patients typically have atrophic adrenals and gonads.⁴⁸ Less than 40 cases have been reported.

Similar to nonclassic lipoid CAH, nonclassic SCC deficiency has been described with delayed adrenal insufficiency onset and variable gonadal effect, caused by mutations corresponding to 7-30% of retained enzyme activity.⁵⁵⁻⁵⁸

Since StAR and SCC deficiency are similar clinically and biochemically, DNA testing is the only definitive method to distinguish between the two, with StAR deficiency being more common.

Diagnosis

Neonatal screening for 21OH deficiency is performed by measuring 17OHP in dried blood spots on filter paper. Second-tier screening with LC-MS/MS can efficiently measure a panel of steroids; this has been used to successfully diagnose 11 β OH deficiency,⁵⁹ but the focus of neonatal screening remains the detection of 21OH deficiency. Premature, stressed, or ill infants could have falsely elevated 17OHP; specificity is improved with gestational age stratification.⁶⁰ Testing using 21-deoxycortisol, which is elevated in 21OH deficiency, might increase neonatal screening specificity.⁶¹

If an infant has a positive neonatal screen or is clinically suspicious for CAH (i.e. ambiguous genitalia), confirmatory testing is indicated. Although a basal panel of LC-MS/MS steroids may be diagnostic, diagnosis often requires cosyntropin testing,^{20,62} and is based on a characteristic rise in adrenal hormone(s) preceding the enzymatic block (Table 2). It should be kept in mind that 17OHP may be elevated in other types of CAH, such as 11 β OH or POR deficiency. An alternative approach is urinary steroid profiling, which illustrates the entire steroid metabolome.⁶³ Additional testing and genotyping may be needed to confirm the diagnosis.

Screening for 21OH deficiency after infancy relies on measuring early morning (before 8 am) 17OHP. A level above 1,000 ng/dL (30 nmol/L) is diagnostic for 21OH deficiency, although a random level of 10,000 ng/dL (303 nmol/L) or greater is commonly observed in the classic form (Table 2).²⁰ A level of less than 200 ng/dL (6 nmol/L) usually excludes nonclassic CAH if measured during the follicular phase of a reproductive-age woman.⁶⁴ Cosyntropin stimulation testing is often needed for diagnosis of the nonclassic form.

The diagnosis of POR can be best made with a urinary steroid profile, revealing characteristic precursor accumulation that can be captured by steroid ratios³² while serum steroid analysis

often yields confusing results due to the overlapping effects of combined 17OH and 21OH deficiency. Further criteria for urinary and serum metabolites have been suggested to diagnose POR prenatally or differentiate POR from 21OH deficiency.^{65,66}

Management of CAH

Medical Treatment

Glucocorticoid therapy

The mainstay of treatment in the classic forms of CAH is chronic glucocorticoid therapy (Panel). Because of their growth suppressing effect, long-acting glucocorticoids are avoided in children, but sometimes used in adults.⁶⁸ The goal is to optimize control of excess hormones, while replacing deficient hormones and avoiding the potential Cushingoid side effects of glucocorticoid therapy. Laboratory results should guide but not define management; clinical evaluation should always be considered.

In general, higher doses of glucocorticoids are needed to achieve adequate suppression of hormone excess (i.e. androgens in the virilizing forms of CAH, classic 21OH, 11 β OH; DOC in 17OH deficiency), whereas lower replacement doses of glucocorticoids should suffice if all steroids are deficient. Lower glucocorticoid doses may also be used in the nonclassic forms of CAH, if treatment is indicated.⁶⁹

For women planning to conceive, a glucocorticoid that does not reach the fetus and is inactivated by placental 11 β HSD2, such as prednisone, prednisolone or hydrocortisone, is typically used and continued throughout pregnancy.

Patients with nonclassic CAH are treated according to symptomatology. Children with nonclassic 21OH deficiency should be treated if they have progressive signs and symptoms of virilization with advanced skeletal maturation. Women with nonclassic CAH with signs of androgen excess can often be successfully treated with oral contraceptive, if needed in combination with spironolactone. Glucocorticoid therapy is used for female infertility in nonclassic 21OH deficiency and has been reported to reduce miscarriage rate when taken throughout pregnancy.^{70,71}

Mineralocorticoid replacement

Mineralocorticoid, in the form of fludrocortisone, is given to achieve a plasma renin activity in the normal range in the salt-wasting forms of CAH (Panel)²⁰. The dose is independent of body size, although higher doses are usually needed during the first 6 months of life due to neonatal physiological mineralocorticoid resistance. Infants also require salt supplementation. Although patients with SV 21OH deficiency have some aldosterone production, relative aldosterone insufficiency exists and fludrocortisone is recommended as it allows for glucocorticoid reduction leading to improved height outcomes.⁷²

Glucocorticoid stress dosing

Patients receiving glucocorticoid treatment, including those with the nonclassic form, need to be educated on adrenal crisis prevention and increasing glucocorticoid dose during intercurrent illness (Panel). Intramuscular, subcutaneous, or intravenous hydrocortisone should be given when oral intake is not possible, and stress dose coverage is identical to that recommended in

primary adrenal insufficiency.⁶⁷ Patients with classic 21OH deficiency also have epinephrine deficiency, due to abnormal adrenomedullary formation;⁷³ this places patients at risk for hypoglycemia, especially when fasting, or during acute illness. Adrenomedullary function has not been studied in the rarer forms of CAH.

Patients with nonclassic CAH may have suboptimal cortisol response on stimulation testing (< 18 ug/dl). If asymptomatic, daily glucocorticoid is not indicated, but glucocorticoid stress coverage should be employed during serious illness or major surgery.

Sex steroids

Sex steroid replacement is started around the time of physiological puberty in 17OH deficiency, 3βHSD deficiency, lipoid CAH, SCC deficiency, and POR (as needed). Androgen (males) and estrogen (females), with progestin to induce cyclical bleeding (if uterus present) are advanced slowly to adult regimens (Panel).

,

Anti-hypertensive

In both 11 βOH and 17OH deficiency, glucocorticoid therapy is often sufficient to control hypertension by suppressing DOC. However, because high dose glucocorticoid therapy and complete suppression of the HPA axis should be avoided, DOC is not fully suppressed and many patients eventually become hypertensive. A mineralocorticoid receptor antagonist or calcium channel blocker can be used^{31,74} (Panel).

Controversial Therapies

Genital Surgery

Surgery for disorders of sex development (DSD) is a complex issue that has generated much controversy. Historically, surgeons have recommended surgery based on genital appearance and fertility potential. In the past two decades, some advocacy groups and physicians have recommended delaying surgery so that patients can participate in the decision regarding surgical intervention. Conversely, others have expressed concern regarding the lack of outcome data and psychosocial stress of not doing early surgery. Most importantly, the family should always be educated on the pros and cons of having and not having surgery. An interdisciplinary team of specialists is often required to navigate the decision making process.⁷⁵

An international group of experts in CAH appointed by the Endocrine Society to develop Clinical Practice Guidelines for CAH due to 21OH deficiency concluded that surgery be considered for significantly virilized 46,XX CAH patients.²⁰ Timing of the surgery is beyond the scope of this article but options include a one stage approach with neurovascular-sparing clitoroplasty, labioplasty, and vaginoplasty being done simultaneously in infancy (norm in many countries including US and UK), waiting until puberty for any surgery, and performing labioplasty and clitoroplasty in infancy with vaginoplasty delayed until puberty. Most CAH patients with 21OH deficiency prefer early surgery.⁷⁶ Although *in utero* exposure to androgen has been shown to affect behavior with male typical behavior patterns commonly seen in classic 46,XX CAH patients, gender dysphoria is extremely rare and the recommended sex assignment of 46,XX DSD due to CAH is female.⁷⁵

The main challenge of surgery of the 46,XX virilized CAH patient remains the imperfect functional and cosmetic outcomes, including urinary incontinence, vaginal stenosis, and clitoral

pain, all of which can impact psychosocial and sexual well-being.⁷⁷ Many of the new surgical approaches have not been present for long enough to assess outcomes. Patients should be referred to a specialist surgeon with DSD management experience.

Surgical reconstruction of 46,XY atypical genital is complex. Chordee repair and surgery for distal hypospadias have high success rates, but proximal hypospadias repair is more challenging with higher complication and reoperation rates. The main complications are urethral stricture, meatal stenosis, urethrocutaneous fistula and glans wings separation.⁷⁸

Early gonadal neoplastic changes were observed histologically as early as 1 year of age in a 46,XY patient with classic lipoid CAH.⁷⁹ Gonadectomy is recommended in severely affected 46,XY patients being raised female, although risk of gonadal malignancy is unknown.

Prenatal treatment

For over 30 years, dexamethasone was offered to pregnant women at risk for having a child with classic virilizing CAH, aiming to suppress fetal androgen production and reduce virilization of an affected female. Dexamethasone, unlike hydrocortisone and prednisolone, crosses the placental barrier to the fetus without inactivation. Today, prenatal therapy is controversial, as only 1 of 8 fetuses will be an affected female when both parents are carriers. Long-term effects of *in utero* dexamethasone exposure are unknown with potential effects on the brain, behavior and cognition described.⁸⁰⁻⁸³

Testing of fetal cells present in maternal circulation is being studied to avoid 46,XY treatment and initiate early treatment in affected 46,XX.⁸⁴ Cell-free fetal DNA obtained from the mother's plasma as early as 5 weeks gestation has correctly identified fetal CAH status in 14 families.⁸⁵

Multiple international groups, including medical societies in the U.S. and Europe, have stated that prenatal therapy should only be considered in a research setting with full disclosure of the potential risks and benefits.^{20,86} The long-term effects of prenatal dexamethasone exposure requires further study but early non-invasive fetal DNA testing would potentially limit exposure to affected female fetuses.

Bilateral Adrenalectomy

Bilateral adrenalectomy has been successfully used to treat female infertility with uncontrolled hyperandrogenism in 21OH deficiency⁸⁷ and uncontrolled hypertension in 11 β OH deficiency.⁸⁸ Although follow-up of patients who have undergone adrenalectomy is overall positive, patients appear to be at increased risk of adrenal crisis and about one-third develop adrenal rest tissue if glucocorticoid dose is too low.⁸⁹ Adrenalectomy should only be considered in patients who have failed all available medical interventions.

Long-term Complications

Glucocorticoid deficiency is characteristic of the severe forms of CAH, and is potentially life-threatening. A study in Sweden of 588 CAH patients compared to a national population-based registry revealed excess mortality in CAH because of adrenal crises,⁹⁰ highlighting the importance of this aspect of the disease. A cross-sectional questionnaire-based study of 122 CAH patients found the majority of adrenal crises occurred during infancy, with a second peak around late adolescence, and precipitated mainly by respiratory and gastro-intestinal infections.⁹¹

All children with CAH receiving glucocorticoid therapy are at risk for growth impairment and short stature. This effect is dose-dependent, thus management is always aimed at treating with the lowest possible effective dose. Alterations in sex steroid exposure can also influence height. Late puberty may occur with the rare types of CAH associated with sex steroid deficiency, and can enhance adult height; vice versa, exposure to excess androgens and estrogens in the virilizing types of CAH can result in early puberty and early epiphyseal fusion (Figure 3B). A meta-analysis of 35 studies of classic 21OH deficiency showed an average final height of 1.38 standard deviations below the population norm.⁷² Whereas studies reveal an association between higher doses of hydrocortisone and shorter final height,^{92,93} earlier rather than later diagnosis and treatment has been associated with improved height outcomes, emphasizing the importance of prevention of hyperandrogenism.

Cardiovascular disease risk factors commonly coexist with CAH. In a cross-sectional UK study of classic 21OH deficient patients, over half of women were obese; and one-third had insulin resistance.⁹⁴ In a Swedish cohort of 588 CAH patients, there was a higher frequency of hypertension, hyperlipidemia, diabetes, and venous thromboembolism compared to controls.⁹⁵

Long term glucocorticoid exposure, particularly higher doses for achievement of tight control, is a main risk factor for compromising bone health. Studies over the past two decades show evidence of lower BMD in patients with 21OH deficiency as compared to controls, with the prevalence of low BMD (osteoporosis or osteopenia) ranging from 37 to 81%,^{94,96-98} with some studies reporting increased fracture rate.^{96,99}

Both males and females with the hypogonadal forms of CAH suffer from infertility. However, successful pregnancy has been achieved in one woman with classic lipoid CAH with clomiphene

citrate stimulation followed by progesterone supplementation.¹⁰⁰ *In vitro* fertilization and transfer of cryopreserved embryos has successfully resulted in a live birth in lipoid CAH and 17OH deficiency.^{101,102} Patients with POR, *StAR*, and *CYP17A1* mutations may also have ovarian cysts and ovarian cyst torsion.¹⁰³⁻¹⁰⁵

In the virilizing forms of CAH, excess adrenal sex steroids can lead to hypogonadotropic hypogonadism^{106,107} and increased progesterone can interfere with endometrial implantation.^{108,109} This may resolve with optimizing glucocorticoid management and suppression of follicular phase progesterone enhances the likelihood of ovulation and subsequent conception.^{110,111}

A main cause of male infertility in classic 21OH and 11 β OH deficiency is adrenal rest tissue. Adrenal rest is thought to arise from aberrant cells of adrenocortical origin that migrate during fetal development along with the gonad after the adrenal and gonadal cells separate from the urogenital ridge. Adrenal rest is most commonly found in the rete testis (Figure 3C) and has been described in the ovaries and broad ligament.^{112,113} Testicular adrenal rest tissue (TART) causes obstructive azoospermia and deficient spermatogenesis. Low inhibin B levels reflect declining Sertoli cell function and can be used to monitor.¹⁰⁶ The prevalence of TART is between 44-94% of men^{98,107,114,115} and 21-33% of boys^{98,116} with classic 21OH, and has also been reported in 11 β OH and HSD3B2 deficiency,¹¹⁷⁻¹¹⁹ and rarely in nonclassic CAH.¹¹⁴ TART shrinkage and reversal of infertility are possible with glucocorticoid therapy;¹²⁰ however the response is variable, because non-reversible fibrotic changes may occur over time.¹²¹

When unsuccessful, other modalities, such as intra-cytoplasmic sperm injection, could be considered.¹²² Testis-preserving surgery with TART resection has not restored fertility,¹²³ but

can be done for orchialgia; simultaneous successful intraoperative sperm retrieval was recently reported.¹²⁴

Future Directions

Alternative Androgen Synthesis Pathways

The quest for new and improved biomarkers of disease severity or treatment response in CAH has included exploration of alternative androgen synthesis pathways. The so-called “backdoor” pathway, leads to the synthesis of DHT, without DHEA, androstenedione and testosterone as intermediates, originating directly from 17-hydroxyprogesterone (Figure 2A). This pathway has been implicated in the normal development of male genitalia⁵⁴ and the prenatal virilization of affected females.¹⁶ Accumulation of 17-hydroxyprogesterone, as observed in 21OH and POR deficiency, increases the substrate flow to the “backdoor” pathway and subsequent studies have described increased alternative pathway metabolite excretion in patients with CAH due to POR¹²⁵ and 21OHD deficiency.¹²⁶

A further alternative androgen synthesis pathway involves the generation of 11-oxygenated C19 steroids in the adrenal cortex via CYP11B1 activity (Figure 2A),¹²⁷ including, 11-ketotestosterone and 11-keto-DHT, androgens that bind and activate the androgen receptor.^{128,129} 11-oxygenated C19 steroids have been shown to be increased in CAH due to 20H deficiency^{130,131} and exaggerated activity of both backdoor and 11-oxygenated C19 pathways persists in treated patients, even if the classic androgen pathway activity is downregulated.¹³⁰

Insights into these novel steroid markers will help to improve monitoring tools and define treatment targets.

Genetic Advances

Genetic studies of CAH have provided insight into the pathophysiology and subtle clinical aspects of the disease. Initially described in 1989,¹³² the *TNXB* gene which encodes the tenascin-X protein, a glycoprotein expressed in connective tissue, and its highly homologous pseudogene *TNXA*, flank *CYP21A2* and its pseudogene *CYP21A1P*, respectively. Chimeric genes which impair both the *CYP21A2* and *TNXB* genes were found to explain an unusual observed phenotype of a connective tissue dysplasia consistent with hypermobility-type Ehlers Danlos syndrome in patients with 21OH deficiency.^{133,134} This novel syndrome, CAH-X, was prevalent in 8.5 % of a cohort of 246 unrelated patients with 21OH deficiency.¹³⁵

Apart from CAH genes, other genes may modify steroid action, salt balance or androgen sensitivity and affect phenotype.^{136,137,138}

Genotyping is essential in confirming the carrier state, and is useful for genetic counseling or establishing the diagnosis of a patient who cannot undergo accurate hormonal testing due to glucocorticoid therapy. Genotyping may one day predict future outcomes and be efficacious in screening programs.¹³⁹

Novel Therapies

Most of the adverse outcomes in patients with CAH are attributable to hormonal imbalances or treatment related comorbidities. New and improved therapies are being developed that target

different aspects of the pathophysiology of CAH (Figure 4) and are being studied in classic 21OH deficiency.

One approach is to replace cortisol in a physiological manner. Circadian cortisol replacement might achieve improved ACTH control and thus adrenal steroid secretion. A modified-release oral hydrocortisone preparation, Chronocort[®], was successful in achieving lower androgen levels while decreasing the hydrocortisone equivalent dose using a twice-daily regimen in a Phase 2 study of 16 patients with classic 21OH deficiency.¹⁴⁰ A phase 3 study is currently underway (NCT02716818). Continuous subcutaneous hydrocortisone infusion via an insulin pump mimicking cortisol circadian rhythm, has similarly shown adequate ACTH suppression with lower total hydrocortisone dose compared to conventional treatment,¹³⁹⁻¹⁴² and showed improved quality-of-life and fatigue in 8 patients with classic 21-OH deficiency.¹⁴² Long-standing comorbidities, such as insulin resistance and TART, remained mostly unchanged by 6 months, suggesting that early intervention is key and other approaches may be needed to treat well established comorbidities.

As ACTH is the primary driver for excess steroid accumulation, strategies for reducing ACTH are under way. A phase I, proof-of-principle study with a corticotropin-releasing factor type 1 receptor antagonist, lowered morning ACTH or 17OHP in 6 of 8 females with classic 21-OH deficiency after a single dose.¹⁴³ Future, multi-dose trials are needed.

Pharmacologic blockade or inhibition of sex steroid synthesis in prepubertal children or women receiving sex hormone replacement would allow for lower dose glucocorticoid replacement in the virilizing forms of CAH. This was studied in 28 prepubertal children with classic 21OH deficiency using an anti-androgen and aromatase-inhibitor in combination with

lower dose hydrocortisone and fludrocortisone, and was successful in normalizing growth over 2 years.¹⁴⁴ Pharmacologic inhibition of sex steroid synthesis was also tested in adult women with CAH receiving gonadal hormone replacement in a phase I, 6-day, dose-escalation study of Abiraterone acetate, a potent CYP17A1 inhibitor.¹⁴⁵ Androstenedione normalized in 5 of 6 women when abiraterone was added to physiologic doses of glucocorticoid and fludrocortisone, showing promising results. Pharmacologic inhibition of sex steroid synthesis is also being studied with ATR-101 (NCT02804178), an inhibitor of acyl-coenzyme A: cholesterol-O-acyltransferase 1, in a phase II study of classic 21-OH deficiency.¹⁴⁶

Adrenal enzyme inhibitors with adrenolytic properties may be useful in the treatment of CAH. Mitotane, inhibits CYP11B1 and CYP11A1, and is adrenolytic with longer term use. Mitotane was successfully used to shrink TART and restore fertility in a 29-year-old male with classic 21OH deficiency.¹⁴⁷ However, due to multiple toxicities of mitotane, the development of alternative adrenolytic therapies is needed.

CAH is a monogenic disease, therefore gene therapy, with the use of cell-based and gene-editing technologies, may be able to restore the defective steroidogenesis.¹⁴⁸ Adrenal transplantation with novel technology using bovine adrenocortical cells has been successful in animal models of adrenal insufficiency.¹⁴⁹ Future technological and genetic advances may enable us to one day cure CAH.

Conclusion

CAH is a group of rare diseases that if left undiagnosed and untreated would result in high morbidity and mortality. The identification of alternative adrenal biomarkers has provided

insight into the origin and synthesis of steroid production and has the potential to alter disease management. Decades of progress in understanding the genetics and pathophysiology of the various forms of CAH have led to a recent explosion in the investigation of new and improved therapies which promise to improve patient outcome.

Competing Interest Statement

Diala El-Maouche reports no conflict of interest. Dr. Wiebke Arlt served as a scientific consultant to and received research funds from Diurnal Limited. Dr. Deborah P. Merke received research funds from Diurnal Limited and Millendo Therapeutics through the National Institutes of Health Cooperative Research and Development Agreement.

1. Delle Piane L, Rinaudo PF, Miller WL. 150 years of congenital adrenal hyperplasia: translation and commentary of De Crecchio's classic paper from 1865. *Endocrinology* 2015; **156**(4): 1210-7.
2. Eberlein WR, Bongiovanni AM. Partial characterization of urinary adrenocortical steroids in adrenal hyperplasia. *The Journal of clinical investigation* 1955; **34**(8): 1337-43.
3. Eberlein WR, Bongiovanni AM. Congenital adrenal hyperplasia with hypertension: unusual steroid pattern in blood and urine. *The Journal of clinical endocrinology and metabolism* 1955; **15**(12): 1531-4.
4. Prader A, Anders GJ. [On the genetics of congenital lipid hyperplasia of the adrenals]. *Helvetica paediatrica acta* 1962; **17**: 285-9.
5. Bongiovanni AM. The Adrenogenital Syndrome with Deficiency of 3 β -hydroxysteroid Dehydrogenase. *The Journal of clinical investigation* 1962; **41**(11): 2086-92.
6. Biglieri EG, Herron MA, Brust N. 17-hydroxylation deficiency in man. *The Journal of clinical investigation* 1966; **45**(12): 1946-54.
7. Pang S, Murphey W, Levine LS, et al. A pilot newborn screening for congenital adrenal hyperplasia in Alaska. *The Journal of clinical endocrinology and metabolism* 1982; **55**(3): 413-20.
8. Peterson RE, Imperato-McGinley J, Gautier T, Shackleton C. Male pseudohermaphroditism due to multiple defects in steroid-biosynthetic microsomal mixed-function oxidases. A new variant of congenital adrenal hyperplasia. *The New England journal of medicine* 1985; **313**(19): 1182-91.
9. White PC, Grossberger D, Onufer BJ, et al. Two genes encoding steroid 21-hydroxylase are located near the genes encoding the fourth component of complement in man. *Proceedings of the National Academy of Sciences of the United States of America* 1985; **82**(4): 1089-93.
10. Kagimoto M, Winter JS, Kagimoto K, Simpson ER, Waterman MR. Structural characterization of normal and mutant human steroid 17 alpha-hydroxylase genes: molecular basis of one example of combined 17 alpha-hydroxylase/17,20 lyase deficiency. *Molecular endocrinology (Baltimore, Md)* 1988; **2**(6): 564-70.
11. White PC, Dupont J, New MI, Leiberman E, Hochberg Z, Rosler A. A mutation in CYP11B1 (Arg-448----His) associated with steroid 11 beta-hydroxylase deficiency in Jews of Moroccan origin. *The Journal of clinical investigation* 1991; **87**(5): 1664-7.
12. Rheaume E, Simard J, Morel Y, et al. Congenital adrenal hyperplasia due to point mutations in the type II 3 beta-hydroxysteroid dehydrogenase gene. *Nature genetics* 1992; **1**(4): 239-45.
13. Lin D, Sugawara T, Strauss JF, 3rd, et al. Role of steroidogenic acute regulatory protein in adrenal and gonadal steroidogenesis. *Science (New York, NY)* 1995; **267**(5205): 1828-31.
14. Tajima T, Fujieda K, Kouda N, Nakae J, Miller WL. Heterozygous mutation in the cholesterol side chain cleavage enzyme (p450scc) gene in a patient with 46,XY sex reversal and adrenal insufficiency. *The Journal of clinical endocrinology and metabolism* 2001; **86**(8): 3820-5.
15. Fluck CE, Tajima T, Pandey AV, et al. Mutant P450 oxidoreductase causes disordered steroidogenesis with and without Antley-Bixler syndrome. *Nature genetics* 2004; **36**(3): 228-30.
16. Arlt W, Walker EA, Draper N, et al. Congenital adrenal hyperplasia caused by mutant P450 oxidoreductase and human androgen synthesis: analytical study. *Lancet (London, England)* 2004; **363**(9427): 2128-35.

17. Bartter FC, Albright F, Forbes AP, Leaf A, Dempsey E, Carroll E. The effects of adrenocorticotrophic hormone and cortisone in the adrenogenital syndrome associated with congenital adrenal hyperplasia: an attempt to explain and correct its disordered hormonal pattern. *The Journal of clinical investigation* 1951; **30**(3): 237-51.
18. Bongiovanni AM, Root AW. The Adrenogenital Syndrome. *The New England journal of medicine* 1963; **268**: 1283-9; 342-51; 391-9.
19. Wilkins L, Lewis RA, Klein R, et al. Treatment of congenital adrenal hyperplasia with cortisone. *The Journal of clinical endocrinology and metabolism* 1951; **11**(1): 1-25.
20. Speiser PW, Azziz R, Baskin LS, et al. Congenital adrenal hyperplasia due to steroid 21-hydroxylase deficiency: an Endocrine Society clinical practice guideline. *The Journal of clinical endocrinology and metabolism* 2010; **95**(9): 4133-60.
21. Coulm B, Coste J, Tardy V, et al. Efficiency of neonatal screening for congenital adrenal hyperplasia due to 21-hydroxylase deficiency in children born in mainland France between 1996 and 2003. *Archives of pediatrics & adolescent medicine* 2012; **166**(2): 113-20.
22. Therrell BL. Newborn screening for congenital adrenal hyperplasia. *Endocrinology and metabolism clinics of North America* 2001; **30**(1): 15-30.
23. New MI. Extensive clinical experience: nonclassical 21-hydroxylase deficiency. *The Journal of clinical endocrinology and metabolism* 2006; **91**(11): 4205-14.
24. Finkelstein GP, Chen W, Mehta SP, et al. Comprehensive genetic analysis of 182 unrelated families with congenital adrenal hyperplasia due to 21-hydroxylase deficiency. *The Journal of clinical endocrinology and metabolism* 2011; **96**(1): E161-72.
25. New MI, Abraham M, Gonzalez B, et al. Genotype-phenotype correlation in 1,507 families with congenital adrenal hyperplasia owing to 21-hydroxylase deficiency. *Proceedings of the National Academy of Sciences of the United States of America* 2013; **110**(7): 2611-6.
26. Fluck CE, Meyer-Boni M, Pandey AV, et al. Why boys will be boys: two pathways of fetal testicular androgen biosynthesis are needed for male sexual differentiation. *American journal of human genetics* 2011; **89**(2): 201-18.
27. Miller WL, Auchus RJ. The molecular biology, biochemistry, and physiology of human steroidogenesis and its disorders. *Endocr Rev* 2011; **32**(1): 81-151.
28. White PC, Curnow KM, Pascoe L. Disorders of steroid 11 beta-hydroxylase isozymes. *Endocr Rev* 1994; **15**(4): 421-38.
29. Paperna T, Gershoni-Baruch R, Badarneh K, Kasinetz L, Hochberg Z. Mutations in CYP11B1 and congenital adrenal hyperplasia in Moroccan Jews. *The Journal of clinical endocrinology and metabolism* 2005; **90**(9): 5463-5.
30. Costa-Santos M, Kater CE, Auchus RJ, Brazilian Congenital Adrenal Hyperplasia Multicenter Study G. Two prevalent CYP17 mutations and genotype-phenotype correlations in 24 Brazilian patients with 17-hydroxylase deficiency. *The Journal of clinical endocrinology and metabolism* 2004; **89**(1): 49-60.
31. Auchus RJ. Steroid 17-hydroxylase and 17,20-lyase deficiencies, genetic and pharmacologic. *The Journal of steroid biochemistry and molecular biology* 2016; **165**(Part A): 71-8.
32. Krone N, Reisch N, Idkowiak J, et al. Genotype-phenotype analysis in congenital adrenal hyperplasia due to P450 oxidoreductase deficiency. *The Journal of clinical endocrinology and metabolism* 2012; **97**(2): E257-67.

33. Bose HS, Sato S, Aisenberg J, Shalev SA, Matsuo N, Miller WL. Mutations in the steroidogenic acute regulatory protein (StAR) in six patients with congenital lipoid adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2000; **85**(10): 3636-9.
34. Guran T, Buonocore F, Saka N, et al. Rare Causes of Primary Adrenal Insufficiency: Genetic and Clinical Characterization of a Large Nationwide Cohort. *The Journal of clinical endocrinology and metabolism* 2016; **101**(1): 284-92.
35. White PC, Vitek A, Dupont B, New MI. Characterization of frequent deletions causing steroid 21-hydroxylase deficiency. *Proceedings of the National Academy of Sciences of the United States of America* 1988; **85**(12): 4436-40.
36. Nandagopal R, Sinaii N, Avila NA, et al. Phenotypic profiling of parents with cryptic nonclassic congenital adrenal hyperplasia: findings in 145 unrelated families. *European journal of endocrinology / European Federation of Endocrine Societies* 2011; **164**(6): 977-84.
37. Khattab A, Haider S, Kumar A, et al. Clinical, genetic, and structural basis of congenital adrenal hyperplasia due to 11beta-hydroxylase deficiency. *Proceedings of the National Academy of Sciences of the United States of America* 2017; **114**(10): E1933-E40.
38. White PC. Congenital adrenal hyperplasia owing to 11beta-hydroxylase deficiency. *Advances in experimental medicine and biology* 2011; **707**: 7-8.
39. Reisch N, Hogler W, Parajes S, et al. A diagnosis not to be missed: nonclassic steroid 11beta-hydroxylase deficiency presenting with premature adrenarche and hirsutism. *The Journal of clinical endocrinology and metabolism* 2013; **98**(10): E1620-5.
40. Geller DH, Auchus RJ, Mendonca BB, Miller WL. The genetic and functional basis of isolated 17,20-lyase deficiency. *Nature genetics* 1997; **17**(2): 201-5.
41. Idkowiak J, Randell T, Dhir V, et al. A missense mutation in the human cytochrome b5 gene causes 46,XY disorder of sex development due to true isolated 17,20 lyase deficiency. *The Journal of clinical endocrinology and metabolism* 2012; **97**(3): E465-75.
42. Chang YT, Zhang L, Alkaddour HS, et al. Absence of molecular defect in the type II 3 beta-hydroxysteroid dehydrogenase (3 beta-HSD) gene in premature pubarche children and hirsute female patients with moderately decreased adrenal 3 beta-HSD activity. *Pediatric research* 1995; **37**(6): 820-4.
43. Mermejo LM, Elias LL, Marui S, Moreira AC, Mendonca BB, de Castro M. Refining hormonal diagnosis of type II 3beta-hydroxysteroid dehydrogenase deficiency in patients with premature pubarche and hirsutism based on HSD3B2 genotyping. *The Journal of clinical endocrinology and metabolism* 2005; **90**(3): 1287-93.
44. Pang S. The molecular and clinical spectrum of 3beta-hydroxysteroid dehydrogenase deficiency disorder. *Trends in endocrinology and metabolism: TEM* 1998; **9**(2): 82-6.
45. Tomalik-Scharte D, Maiter D, Kirchheiner J, Ivison HE, Fuhr U, Arlt W. Impaired hepatic drug and steroid metabolism in congenital adrenal hyperplasia due to P450 oxidoreductase deficiency. *European journal of endocrinology / European Federation of Endocrine Societies* 2010; **163**(6): 919-24.
46. Laue K, Pogoda HM, Daniel PB, et al. Craniosynostosis and multiple skeletal anomalies in humans and zebrafish result from a defect in the localized degradation of retinoic acid. *American journal of human genetics* 2011; **89**(5): 595-606.

47. Otto DM, Henderson CJ, Carrie D, et al. Identification of novel roles of the cytochrome p450 system in early embryogenesis: effects on vasculogenesis and retinoic Acid homeostasis. *Mol Cell Biol* 2003; **23**(17): 6103-16.
48. Miller WL. Disorders in the initial steps of steroid hormone synthesis. *The Journal of steroid biochemistry and molecular biology* 2016; **165**(Part A): 18-37.
49. Chen X, Baker BY, Abduljabbar MA, Miller WL. A genetic isolate of congenital lipid adrenal hyperplasia with atypical clinical findings. *The Journal of clinical endocrinology and metabolism* 2005; **90**(2): 835-40.
50. Bose HS, Sugawara T, Strauss JF, 3rd, Miller WL, International Congenital Lipoid Adrenal Hyperplasia C. The pathophysiology and genetics of congenital lipid adrenal hyperplasia. *The New England journal of medicine* 1996; **335**(25): 1870-8.
51. Sugawara T, Holt JA, Driscoll D, et al. Human steroidogenic acute regulatory protein: functional activity in COS-1 cells, tissue-specific expression, and mapping of the structural gene to 8p11.2 and a pseudogene to chromosome 13. *Proceedings of the National Academy of Sciences of the United States of America* 1995; **92**(11): 4778-82.
52. Baker BY, Lin L, Kim CJ, et al. Nonclassic congenital lipid adrenal hyperplasia: a new disorder of the steroidogenic acute regulatory protein with very late presentation and normal male genitalia. *The Journal of clinical endocrinology and metabolism* 2006; **91**(12): 4781-5.
53. Metherell LA, Naville D, Halaby G, et al. Nonclassic lipid congenital adrenal hyperplasia masquerading as familial glucocorticoid deficiency. *The Journal of clinical endocrinology and metabolism* 2009; **94**(10): 3865-71.
54. Fluck CE, Pandey AV, Dick B, et al. Characterization of novel StAR (steroidogenic acute regulatory protein) mutations causing non-classic lipid adrenal hyperplasia. *PloS one* 2011; **6**(5): e20178.
55. Rubtsov P, Karmanov M, Sverdlova P, Spirin P, Tiulpakov A. A novel homozygous mutation in CYP11A1 gene is associated with late-onset adrenal insufficiency and hypospadias in a 46,XY patient. *The Journal of clinical endocrinology and metabolism* 2009; **94**(3): 936-9.
56. Sahakitrungruang T, Tee MK, Blackett PR, Miller WL. Partial defect in the cholesterol side-chain cleavage enzyme P450_{scc} (CYP11A1) resembling nonclassic congenital lipid adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2011; **96**(3): 792-8.
57. Tee MK, Abramsohn M, Loewenthal N, et al. Varied clinical presentations of seven patients with mutations in CYP11A1 encoding the cholesterol side-chain cleavage enzyme, P450_{scc}. *The Journal of clinical endocrinology and metabolism* 2013; **98**(2): 713-20.
58. Parajes S, Kamrath C, Rose IT, et al. A novel entity of clinically isolated adrenal insufficiency caused by a partially inactivating mutation of the gene encoding for P450 side chain cleavage enzyme (CYP11A1). *The Journal of clinical endocrinology and metabolism* 2011; **96**(11): E1798-806.
59. Janzen N, Riepe FG, Peter M, et al. Neonatal screening: identification of children with 11beta-hydroxylase deficiency by second-tier testing. *Horm Res Paediatr* 2012; **77**(3): 195-9.
60. van der Kamp HJ, Oudshoorn CG, Elvers BH, et al. Cutoff levels of 17-alpha-hydroxyprogesterone in neonatal screening for congenital adrenal hyperplasia should be based on gestational age rather than on birth weight. *The Journal of clinical endocrinology and metabolism* 2005; **90**(7): 3904-7.

61. Janzen N, Peter M, Sander S, et al. Newborn screening for congenital adrenal hyperplasia: additional steroid profile using liquid chromatography-tandem mass spectrometry. *The Journal of clinical endocrinology and metabolism* 2007; **92**(7): 2581-9.
62. New MI, Lorenzen F, Lerner AJ, et al. Genotyping steroid 21-hydroxylase deficiency: hormonal reference data. *The Journal of clinical endocrinology and metabolism* 1983; **57**(2): 320-6.
63. Caulfield MP, Lynn T, Gottschalk ME, et al. The diagnosis of congenital adrenal hyperplasia in the newborn by gas chromatography/mass spectrometry analysis of random urine specimens. *The Journal of clinical endocrinology and metabolism* 2002; **87**(8): 3682-90.
64. Azziz R, Hincapie LA, Knochenhauer ES, Dewailly D, Fox L, Boots LR. Screening for 21-hydroxylase-deficient nonclassic adrenal hyperplasia among hyperandrogenic women: a prospective study. *Fertil Steril* 1999; **72**(5): 915-25.
65. Koyama Y, Homma K, Fukami M, et al. Two-step biochemical differential diagnosis of classic 21-hydroxylase deficiency and cytochrome P450 oxidoreductase deficiency in Japanese infants by GC-MS measurement of urinary pregnanetriolone/ tetrahydrocortisone ratio and 11beta-hydroxyandrosterone. *Clin Chem* 2012; **58**(4): 741-7.
66. Reisch N, Idkowiak J, Hughes BA, et al. Prenatal diagnosis of congenital adrenal hyperplasia caused by P450 oxidoreductase deficiency. *J Clin Endocrinol Metab* 2013; **98**(3): E528-36.
67. Bornstein SR, Allolio B, Arlt W, et al. Diagnosis and Treatment of Primary Adrenal Insufficiency: An Endocrine Society Clinical Practice Guideline. *The Journal of clinical endocrinology and metabolism* 2016; **101**(2): 364-89.
68. Auchus RJ, Arlt W. Approach to the patient: the adult with congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2013; **98**(7): 2645-55.
69. Livadas S, Dracopoulou M, Dastamani A, et al. The spectrum of clinical, hormonal and molecular findings in 280 individuals with nonclassical congenital adrenal hyperplasia caused by mutations of the CYP21A2 gene. *Clinical endocrinology* 2015; **82**(4): 543-9.
70. Bidet M, Bellanne-Chantelot C, Galand-Portier MB, et al. Fertility in women with nonclassical congenital adrenal hyperplasia due to 21-hydroxylase deficiency. *The Journal of clinical endocrinology and metabolism* 2010; **95**(3): 1182-90.
71. Trakakis E, Dracopoulou-Vabouli M, Dacou-Voutetakis C, Basios G, Chrelias C, Kassanos D. Infertility reversed by glucocorticoids and full-term pregnancy in a couple with previously undiagnosed nonclassic congenital adrenal hyperplasia. *Fertil Steril* 2011; **96**(4): 1048-50.
72. Muthusamy K, Elamin MB, Smushkin G, et al. Clinical review: Adult height in patients with congenital adrenal hyperplasia: a systematic review and metaanalysis. *The Journal of clinical endocrinology and metabolism* 2010; **95**(9): 4161-72.
73. Merke DP, Chrousos GP, Eisenhofer G, et al. Adrenomedullary dysplasia and hypofunction in patients with classic 21-hydroxylase deficiency. *N Engl J Med* 2000; **343**(19): 1362-8.
74. Bulsari K, Falhammar H. Clinical perspectives in congenital adrenal hyperplasia due to 11beta-hydroxylase deficiency. *Endocrine* 2017; **55**(1): 19-36.
75. Fisher AD, Ristori J, Fanni E, Castellini G, Forti G, Maggi M. Gender identity, gender assignment and reassignment in individuals with disorders of sex development: a major of dilemma. *Journal of endocrinological investigation* 2016; **39**(11): 1207-24.

76. Nordenskjold A, Holmdahl G, Frisen L, et al. Type of mutation and surgical procedure affect long-term quality of life for women with congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2008; **93**(2): 380-6.
77. Merke DP, Poppas DP. Management of adolescents with congenital adrenal hyperplasia. *Lancet Diabetes Endocrinol* 2013; **1**(4): 341-52.
78. DiSandro M, Merke DP, Rink RC. Review of current surgical techniques and medical management considerations in the treatment of pediatric patients with disorders of sex development. *Hormone and metabolic research = Hormon- und Stoffwechselforschung = Hormones et metabolisme* 2015; **47**(5): 321-8.
79. Abdulhadi-Atwan M, Jean A, Chung WK, et al. Role of a founder c.201_202delCT mutation and new phenotypic features of congenital lipoid adrenal hyperplasia in Palestinians. *J Clin Endocrinol Metab* 2007; **92**(10): 4000-8.
80. Hirvikoski T, Nordenstrom A, Lindholm T, et al. Cognitive functions in children at risk for congenital adrenal hyperplasia treated prenatally with dexamethasone. *The Journal of clinical endocrinology and metabolism* 2007; **92**(2): 542-8.
81. Lajic S, Nordenstrom A, Hirvikoski T. Long-term outcome of prenatal dexamethasone treatment of 21-hydroxylase deficiency. *Endocrine development* 2011; **20**: 96-105.
82. Wallensteen L, Zimmermann M, Sandberg MT, et al. Evaluation of behavioral problems after prenatal dexamethasone treatment in Swedish adolescents at risk of CAH. *Horm Behav* 2016; **85**: 5-11.
83. Wallensteen L, Zimmermann M, Thomsen Sandberg M, et al. Sex-Dimorphic Effects of Prenatal Treatment With Dexamethasone. *The Journal of clinical endocrinology and metabolism* 2016; **101**(10): 3838-46.
84. Tardy-Guidollet V, Menassa R, Costa JM, et al. New management strategy of pregnancies at risk of congenital adrenal hyperplasia using fetal sex determination in maternal serum: French cohort of 258 cases (2002-2011). *The Journal of clinical endocrinology and metabolism* 2014; **99**(4): 1180-8.
85. New MI, Tong YK, Yuen T, et al. Noninvasive prenatal diagnosis of congenital adrenal hyperplasia using cell-free fetal DNA in maternal plasma. *The Journal of clinical endocrinology and metabolism* 2014; **99**(6): E1022-30.
86. Hirvikoski T, Nordenstrom A, Wedell A, Ritzen M, Lajic S. Prenatal dexamethasone treatment of children at risk for congenital adrenal hyperplasia: the Swedish experience and standpoint. *J Clin Endocrinol Metab* 2012; **97**(6): 1881-3.
87. Dagalakis U, Mallappa A, Elman M, Quezado M, Merke DP. Positive fertility outcomes in a female with classic congenital adrenal hyperplasia following bilateral adrenalectomy. *International journal of pediatric endocrinology* 2016; **2016**: 10.
88. Chabre O, Portrat-Doyen S, Chaffanjon P, et al. Bilateral laparoscopic adrenalectomy for congenital adrenal hyperplasia with severe hypertension, resulting from two novel mutations in splice donor sites of CYP11B1. *The Journal of clinical endocrinology and metabolism* 2000; **85**(11): 4060-8.
89. Van Wyk JJ, Ritzen EM. The role of bilateral adrenalectomy in the treatment of congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2003; **88**(7): 2993-8.

90. Falhammar H, Frisen L, Norrby C, et al. Increased mortality in patients with congenital adrenal hyperplasia due to 21-hydroxylase deficiency. *J Clin Endocrinol Metab* 2014; **99**(12): E2715-21.
91. Reisch N, Willige M, Kohn D, et al. Frequency and causes of adrenal crises over lifetime in patients with 21-hydroxylase deficiency. *European journal of endocrinology / European Federation of Endocrine Societies* 2012; **167**(1): 35-42.
92. Cordeiro GV, Silva IN, Goulart EM, Chagas AJ, Kater CE. Final height in congenital adrenal hyperplasia: the dilemma of hypercortisolism versus hyperandrogenism. *Arquivos brasileiros de endocrinologia e metabologia* 2013; **57**(2): 126-31.
93. Sarafoglou K, Addo OY, Turcotte L, et al. Impact of hydrocortisone on adult height in congenital adrenal hyperplasia-the Minnesota cohort. *The Journal of pediatrics* 2014; **164**(5): 1141-6 e1.
94. Arlt W, Willis DS, Wild SH, et al. Health status of adults with congenital adrenal hyperplasia: a cohort study of 203 patients. *The Journal of clinical endocrinology and metabolism* 2010; **95**(11): 5110-21.
95. Falhammar H, Frisen L, Hirschberg AL, et al. Increased Cardiovascular and Metabolic Morbidity in Patients With 21-Hydroxylase Deficiency: A Swedish Population-Based National Cohort Study. *The Journal of clinical endocrinology and metabolism* 2015; **100**(9): 3520-8.
96. El-Maouche D, Collier S, Prasad M, Reynolds JC, Merke DP. Cortical bone mineral density in patients with congenital adrenal hyperplasia due to 21-hydroxylase deficiency. *Clinical endocrinology* 2015; **82**(3): 330-7.
97. Falhammar H, Filipsson Nystrom H, Wedell A, Brismar K, Thoren M. Bone mineral density, bone markers, and fractures in adult males with congenital adrenal hyperplasia. *Eur J Endocrinol* 2013; **168**(3): 331-41.
98. Finkelstein GP, Kim MS, Sinaii N, et al. Clinical characteristics of a cohort of 244 patients with congenital adrenal hyperplasia. *J Clin Endocrinol Metab* 2012; **97**(12): 4429-38.
99. Falhammar H, Filipsson H, Holmdahl G, et al. Fractures and bone mineral density in adult women with 21-hydroxylase deficiency. *The Journal of clinical endocrinology and metabolism* 2007; **92**(12): 4643-9.
100. Khoury K, Barbar E, Ainmelk Y, Ouellet A, Lavigne P, LeHoux JG. Thirty-Eight-Year Follow-Up of Two Sibling Lipoid Congenital Adrenal Hyperplasia Patients Due to Homozygous Steroidogenic Acute Regulatory (STARD1) Protein Mutation. Molecular Structure and Modeling of the STARD1 L275P Mutation. *Front Neurosci* 2016; **10**: 527.
101. Albarel F, Perrin J, Jegaden M, et al. Successful IVF pregnancy despite inadequate ovarian steroidogenesis due to congenital lipoid adrenal hyperplasia (CLAH): a case report. *Hum Reprod* 2016; **31**(11): 2609-12.
102. Bianchi PH, Gouveia GR, Costa EM, et al. Successful Live Birth in a Woman With 17alpha-Hydroxylase Deficiency Through IVF Frozen-Thawed Embryo Transfer. *The Journal of clinical endocrinology and metabolism* 2016; **101**(2): 345-8.
103. Idkowiak J, O'Riordan S, Reisch N, et al. Pubertal presentation in seven patients with congenital adrenal hyperplasia due to P450 oxidoreductase deficiency. *The Journal of clinical endocrinology and metabolism* 2011; **96**(3): E453-62.
104. Jin HY, Choi JH, Lee BH, Kim GH, Kim HK, Yoo HW. Ovarian cyst torsion in a patient with congenital lipoid adrenal hyperplasia. *European journal of pediatrics* 2011; **170**(4): 535-8.

105. Roger M, Merceron RE, Girard F, et al. Dexamethasone-suppressible hypercorticism in two 46,XX subjects with ambiguous genitalia and ovarian cysts. Partial defect of 17 alpha-hydroxylase or 17-20-desmolase. *Hormone research* 1982; **16**(1): 23-31.
106. Reisch N, Flade L, Scherr M, et al. High prevalence of reduced fecundity in men with congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2009; **94**(5): 1665-70.
107. Stikkelbroeck NM, Otten BJ, Pasic A, et al. High prevalence of testicular adrenal rest tumors, impaired spermatogenesis, and Leydig cell failure in adolescent and adult males with congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2001; **86**(12): 5721-8.
108. Labarta E, Martinez-Conejero JA, Alama P, et al. Endometrial receptivity is affected in women with high circulating progesterone levels at the end of the follicular phase: a functional genomics analysis. *Hum Reprod* 2011; **26**(7): 1813-25.
109. Mnif MF, Kamoun M, Kacem FH, et al. Reproductive outcomes of female patients with congenital adrenal hyperplasia due to 21-hydroxylase deficiency. *Indian J Endocrinol Metab* 2013; **17**(5): 790-3.
110. Casteras A, De Silva P, Rumsby G, Conway GS. Reassessing fecundity in women with classical congenital adrenal hyperplasia (CAH): normal pregnancy rate but reduced fertility rate. *Clinical endocrinology* 2009; **70**(6): 833-7.
111. Lekarev O, Lin-Su K, Vogiatzi MG. Infertility and Reproductive Function in Patients with Congenital Adrenal Hyperplasia: Pathophysiology, Advances in Management, and Recent Outcomes. *Endocrinol Metab Clin North Am* 2015; **44**(4): 705-22.
112. Crocker MK, Barak S, Millo CM, et al. Use of PET/CT with cosyntropin stimulation to identify and localize adrenal rest tissue following adrenalectomy in a woman with congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2012; **97**(11): E2084-9.
113. Russo G, Paesano P, Taccagni G, Del Maschio A, Chiumello G. Ovarian adrenal-like tissue in congenital adrenal hyperplasia. *The New England journal of medicine* 1998; **339**(12): 853-4.
114. Falhammar H, Nystrom HF, Ekstrom U, Granberg S, Wedell A, Thoren M. Fertility, sexuality and testicular adrenal rest tumors in adult males with congenital adrenal hyperplasia. *Eur J Endocrinol* 2012; **166**(3): 441-9.
115. Yu MK, Jung MK, Kim KE, et al. Clinical manifestations of testicular adrenal rest tumor in males with congenital adrenal hyperplasia. *Ann Pediatr Endocrinol Metab* 2015; **20**(3): 155-61.
116. Martinez-Aguayo A, Rocha A, Rojas N, et al. Testicular adrenal rest tumors and Leydig and Sertoli cell function in boys with classical congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2007; **92**(12): 4583-9.
117. Aycan Z, Bas VN, Cetinkaya S, Yilmaz Agladioglu S, Tiryaki T. Prevalence and long-term follow-up outcomes of testicular adrenal rest tumours in children and adolescent males with congenital adrenal hyperplasia. *Clinical endocrinology* 2013; **78**(5): 667-72.
118. Chihaoui M, Kanoun F, Chaker F, et al. Testicular adrenal rest tumours in young adult males with congenital adrenal hyperplasia: prevalence and impact on testicular function. *Andrologia* 2016; **48**(1): 45-50.
119. Claahsen-van der Grinten HL, Duthoi K, Otten BJ, d'Ancona FC, Hulsbergen-vd Kaa CA, Hermus AR. An adrenal rest tumour in the perirenal region in a patient with congenital adrenal

- hyperplasia due to congenital 3beta-hydroxysteroid dehydrogenase deficiency. *European journal of endocrinology / European Federation of Endocrine Societies* 2008; **159**(4): 489-91.
120. Stikkelbroeck NM, Hermus AR, Suliman HM, Jager GJ, Otten BJ. Asymptomatic testicular adrenal rest tumours in adolescent and adult males with congenital adrenal hyperplasia: basal and follow-up investigation after 2.6 years. *Journal of pediatric endocrinology & metabolism : JPEM* 2004; **17**(4): 645-53.
121. Claahsen-van der Grinten HL, Otten BJ, Stikkelbroeck MM, Sweep FC, Hermus AR. Testicular adrenal rest tumours in congenital adrenal hyperplasia. *Best practice & research Clinical endocrinology & metabolism* 2009; **23**(2): 209-20.
122. Walker BR, Skoog SJ, Winslow BH, Canning DA, Tank ES. Testis sparing surgery for steroid unresponsive testicular tumors of the adrenogenital syndrome. *The Journal of urology* 1997; **157**(4): 1460-3.
123. Claahsen-van der Grinten HL, Otten BJ, Takahashi S, et al. Testicular adrenal rest tumors in adult males with congenital adrenal hyperplasia: evaluation of pituitary-gonadal function before and after successful testis-sparing surgery in eight patients. *The Journal of clinical endocrinology and metabolism* 2007; **92**(2): 612-5.
124. Kavoussi PK, Summers-Colquitt RB, Odenwald KC, et al. Sperm retrieval and concomitant tumor resection in azoospermic men with congenital adrenal hyperplasia and bilateral testicular adrenal rest tumors: a case report. *J Assist Reprod Genet* 2016; **33**(4): 545-8.
125. Homma K, Hasegawa T, Nagai T, et al. Urine steroid hormone profile analysis in cytochrome P450 oxidoreductase deficiency: implication for the backdoor pathway to dihydrotestosterone. *The Journal of clinical endocrinology and metabolism* 2006; **91**(7): 2643-9.
126. Kamrath C, Hochberg Z, Hartmann MF, Remer T, Wudy SA. Increased activation of the alternative "backdoor" pathway in patients with 21-hydroxylase deficiency: evidence from urinary steroid hormone analysis. *The Journal of clinical endocrinology and metabolism* 2012; **97**(3): E367-75.
127. Pretorius E, Arlt W, Storbeck KH. A new dawn for androgens: Novel lessons from 11-oxygenated C19 steroids. *Molecular and cellular endocrinology* 2016; **441**: 76-85.
128. Pretorius E, Africander DJ, Vlok M, Perkins MS, Quanson J, Storbeck KH. 11-Ketotestosterone and 11-Ketodihydrotestosterone in Castration Resistant Prostate Cancer: Potent Androgens Which Can No Longer Be Ignored. *PLoS one* 2016; **11**(7): e0159867.
129. Storbeck KH, Bloem LM, Africander D, Schloms L, Swart P, Swart AC. 11beta-Hydroxydihydrotestosterone and 11-ketodihydrotestosterone, novel C19 steroids with androgenic activity: a putative role in castration resistant prostate cancer? *Molecular and cellular endocrinology* 2013; **377**(1-2): 135-46.
130. Jones CM, Mallappa A, Reisch N, et al. Modified release and conventional glucocorticoids and diurnal androgen excretion in congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2016: jc20162855.
131. Turcu AF, Nanba AT, Chomic R, et al. Adrenal-derived 11-oxygenated 19-carbon steroids are the dominant androgens in classic 21-hydroxylase deficiency. *European journal of endocrinology / European Federation of Endocrine Societies* 2016; **174**(5): 601-9.
132. Morel Y, Bristow J, Gitelman SE, Miller WL. Transcript encoded on the opposite strand of the human steroid 21-hydroxylase/complement component C4 gene locus. *Proceedings of the National Academy of Sciences of the United States of America* 1989; **86**(17): 6582-6.

133. Burch GH, Gong Y, Liu W, et al. Tenascin-X deficiency is associated with Ehlers-Danlos syndrome. *Nature genetics* 1997; **17**(1): 104-8.
134. Merke DP, Chen W, Morissette R, et al. Tenascin-X haploinsufficiency associated with Ehlers-Danlos syndrome in patients with congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2013; **98**(2): E379-87.
135. Morissette R, Chen W, Perritt AF, et al. Broadening the Spectrum of Ehlers Danlos Syndrome in Patients With Congenital Adrenal Hyperplasia. *The Journal of clinical endocrinology and metabolism* 2015; **100**(8): E1143-52.
136. Gomes LG, Huang N, Agrawal V, Mendonca BB, Bachega TA, Miller WL. Extraadrenal 21-hydroxylation by CYP2C19 and CYP3A4: effect on 21-hydroxylase deficiency. *The Journal of clinical endocrinology and metabolism* 2009; **94**(1): 89-95.
137. Kaupert LC, Lemos-Marini SH, De Mello MP, et al. The effect of fetal androgen metabolism-related gene variants on external genitalia virilization in congenital adrenal hyperplasia. *Clinical genetics* 2013; **84**(5): 482-8.
138. Rocha RO, Billerbeck AE, Pinto EM, et al. The degree of external genitalia virilization in girls with 21-hydroxylase deficiency appears to be influenced by the CAG repeats in the androgen receptor gene. *Clinical endocrinology* 2008; **68**(2): 226-32.
139. White PC. Neonatal screening for congenital adrenal hyperplasia. *Nature reviews Endocrinology* 2009; **5**(9): 490-8.
140. Mallappa A, Sinaii N, Kumar P, et al. A phase 2 study of Chronocort, a modified-release formulation of hydrocortisone, in the treatment of adults with classic congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2015; **100**(3): 1137-45.
141. Bryan SM, Honour JW, Hindmarsh PC. Management of altered hydrocortisone pharmacokinetics in a boy with congenital adrenal hyperplasia using a continuous subcutaneous hydrocortisone infusion. *The Journal of clinical endocrinology and metabolism* 2009; **94**(9): 3477-80.
142. Nella AA, Mallappa A, Perritt AF, et al. A Phase 2 Study of Continuous Subcutaneous Hydrocortisone Infusion in Adults with Congenital Adrenal Hyperplasia. *The Journal of clinical endocrinology and metabolism* 2016: jc20161916.
143. Turcu AF, Spencer-Segal JL, Farber RH, et al. Single-Dose Study of a Corticotropin-Releasing Factor Receptor-1 Antagonist in Women With 21-Hydroxylase Deficiency. *J Clin Endocrinol Metab* 2016; **101**(3): 1174-80.
144. Merke DP, Keil MF, Jones JV, Fields J, Hill S, Cutler GB, Jr. Flutamide, testolactone, and reduced hydrocortisone dose maintain normal growth velocity and bone maturation despite elevated androgen levels in children with congenital adrenal hyperplasia. *The Journal of clinical endocrinology and metabolism* 2000; **85**(3): 1114-20.
145. Auchus RJ, Buschur EO, Chang AY, et al. Abiraterone acetate to lower androgens in women with classic 21-hydroxylase deficiency. *J Clin Endocrinol Metab* 2014; **99**(8): 2763-70.
146. Cheng Y, Kerppola RE, Kerppola TK. ATR-101 disrupts mitochondrial functions in adrenocortical carcinoma cells and in vivo. *Endocrine-related cancer* 2016; **23**(4): 1-19.
147. Bry-Gauillard H, Cartes A, Young J. Mitotane for 21-hydroxylase deficiency in an infertile man. *The New England journal of medicine* 2014; **371**(21): 2042-4.
148. Ruiz-Babot G, Hadjidemetriou I, King PJ, Guasti L. New directions for the treatment of adrenal insufficiency. *Frontiers in endocrinology* 2015; **6**: 70.

149. Balyura M, Gelfgat E, Ehrhart-Bornstein M, et al. Transplantation of bovine adrenocortical cells encapsulated in alginate. *Proceedings of the National Academy of Sciences of the United States of America* 2015; **112**(8): 2527-32.

Figure 1

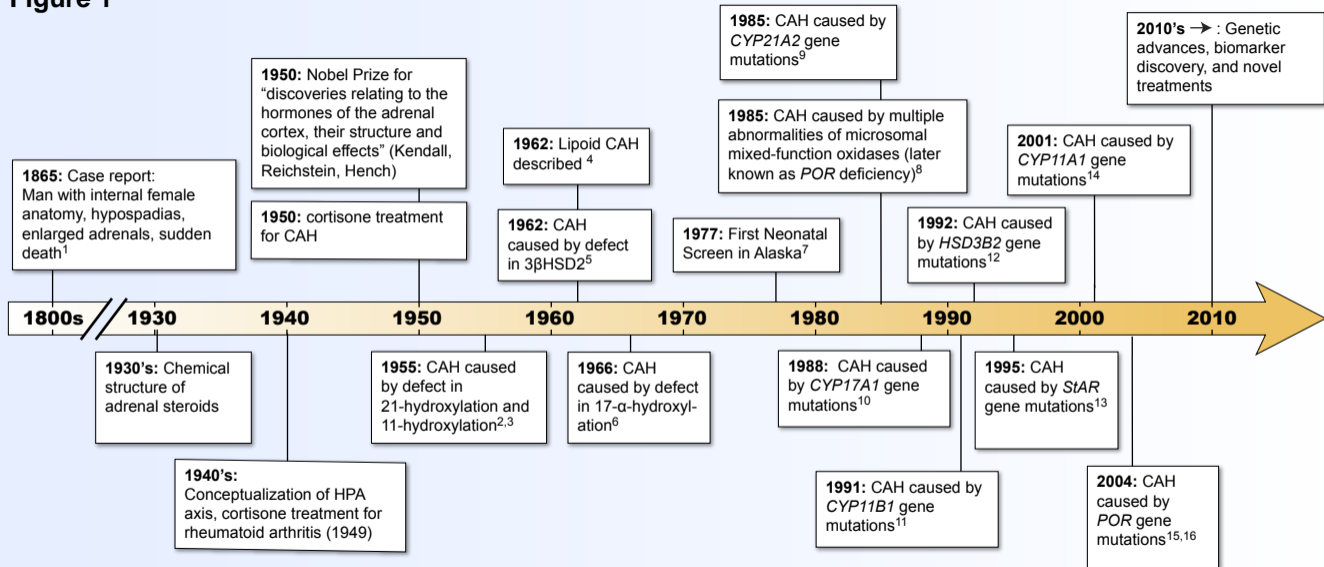
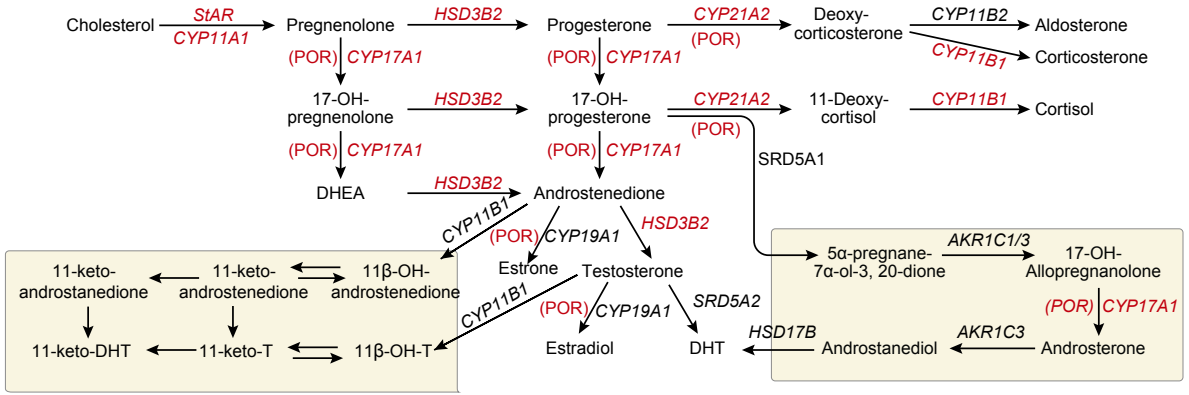
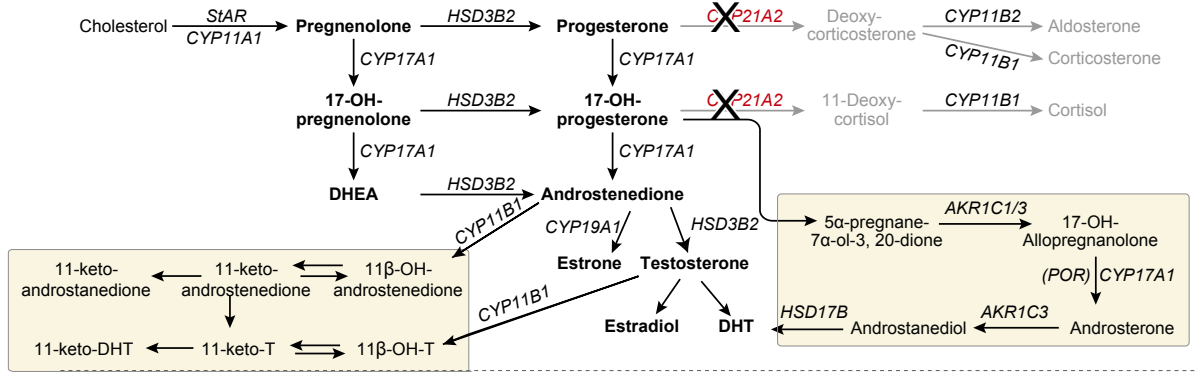
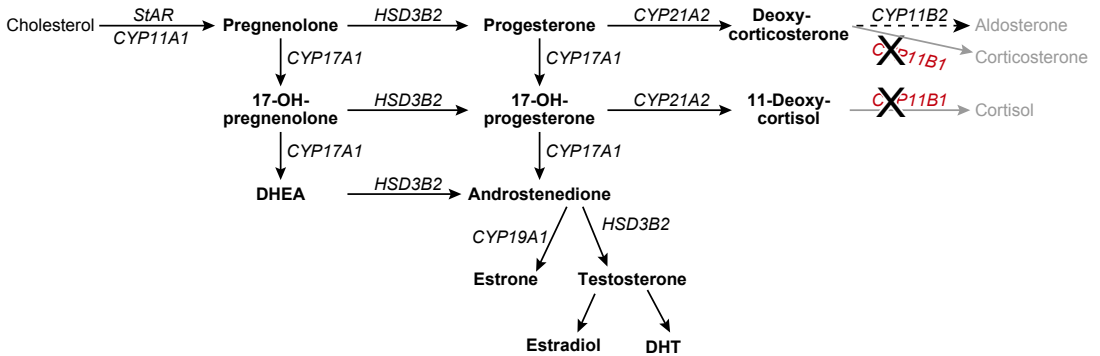
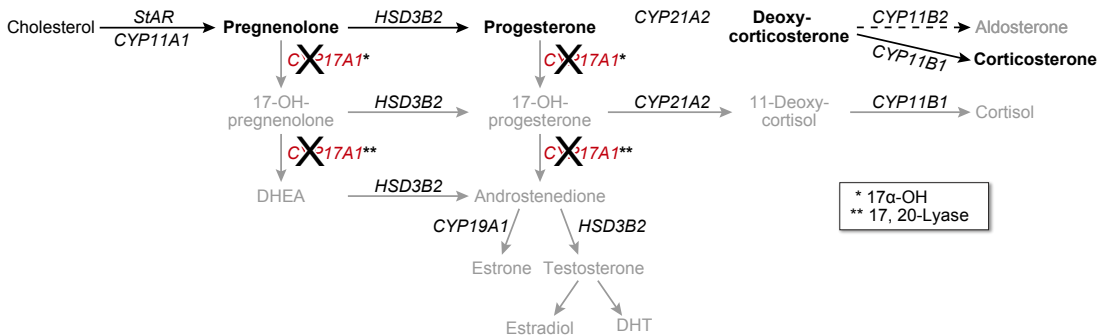
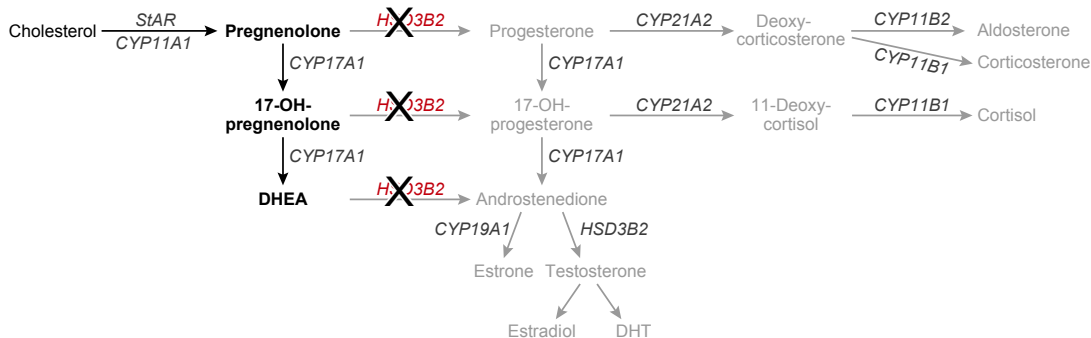
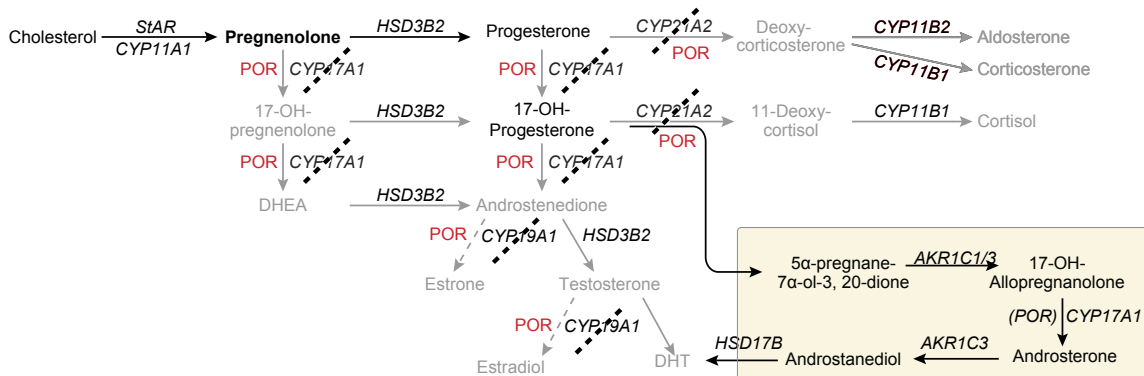


Figure 2**A** Adrenal Hormone Synthesis and Alternative Pathways**B** 21-hydroxylase deficiency**C** 11 β -hydroxylase deficiency**D** 17-hydroxylase deficiency

E 3β HSD2 deficiency



F POR deficiency



G Lipoid congenital adrenal hyperplasia or P450 cholesterol side chain cleavage

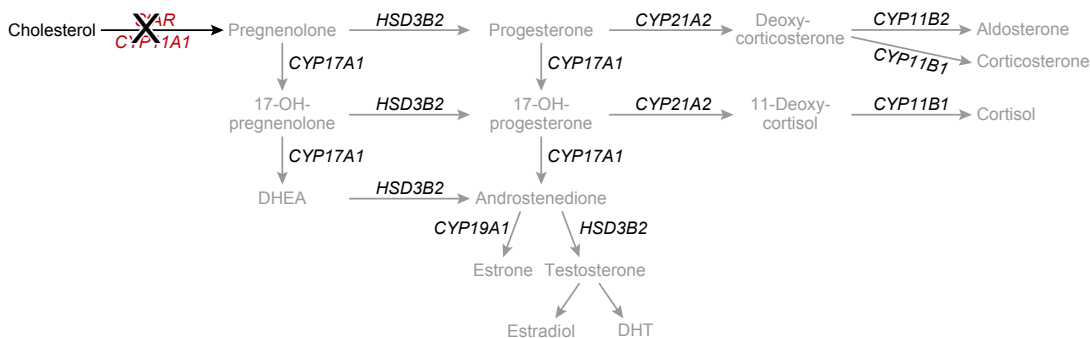


Figure 3A
[Click here to download high resolution image](#)



Figure 3B
 Click here to download high resolution image

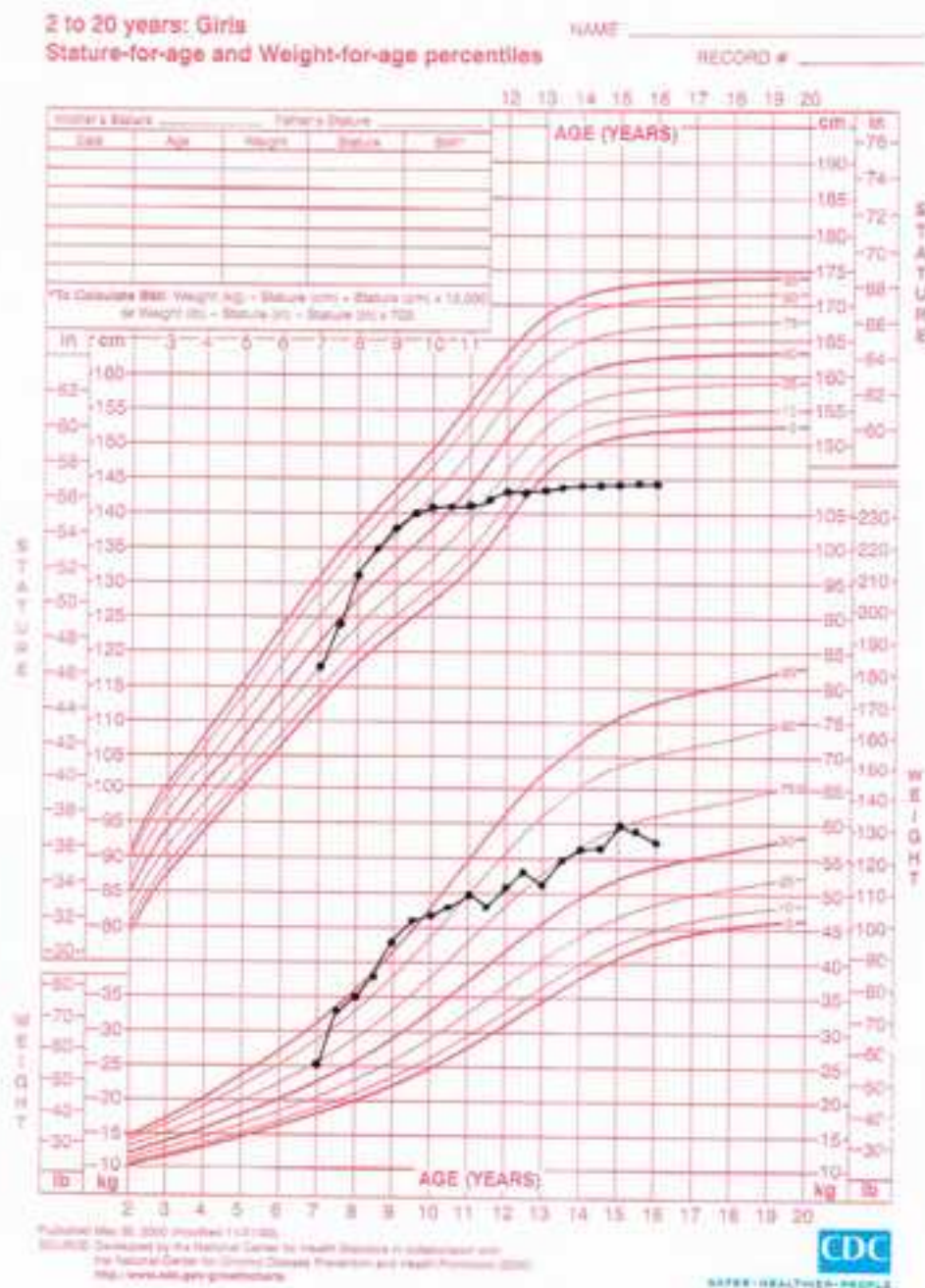


Figure 3C
[Click here to download high resolution image](#)

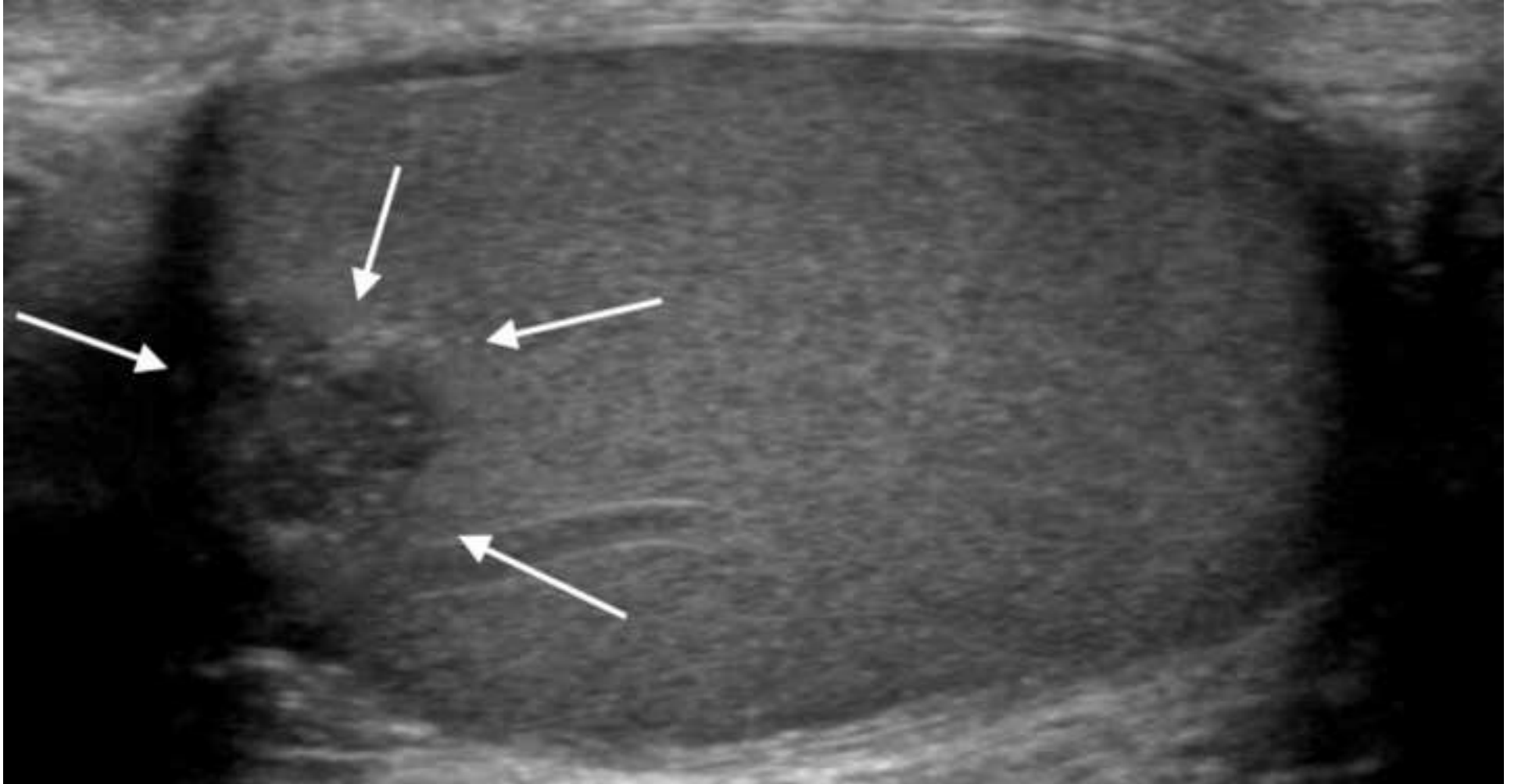


Figure 4

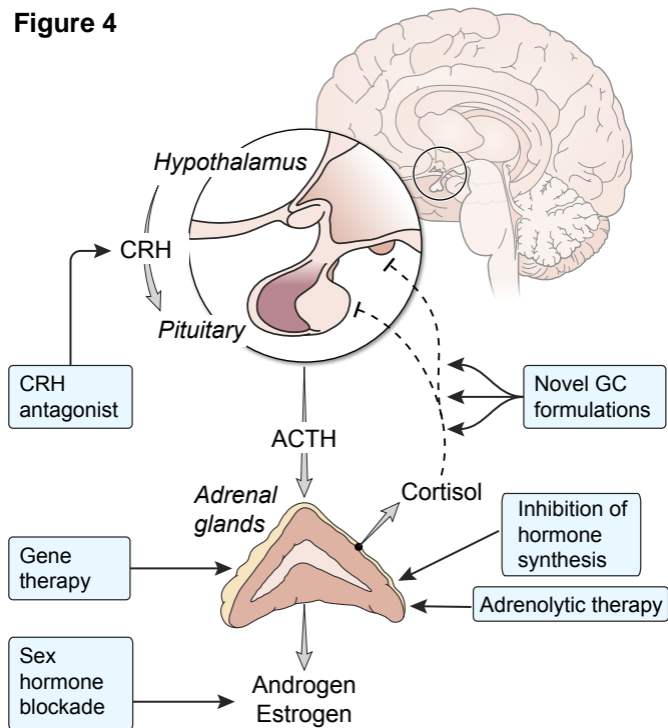


Figure 1: Timeline denoting important discoveries in adrenal steroidogenesis, treatment landmarks and gene discovery of congenital adrenal hyperplasia

Figure 2: Adrenal steroidogenesis pathways

The CAH-causing genes are depicted in red. Panel A shows the classical steroidogenesis pathway and alternative pathways leading to androgen production in the light yellow boxes. Panels B through G show the various forms of CAH and the impact of the specific impairment on the adrenal steroidogenic pathway. Light grey denotes deficient hormones in low levels due to the preceding block in steroid production. Dashed arrows denote indirect suppression of the subsequent hormone. Dashed lines across enzymes denote apparent enzyme deficiency. DHT = dihydrotestosterone; T= Testosterone.

Figure 3: Adverse outcomes in congenital adrenal hyperplasia. Panel A shows atypical genitalia with clitoromegaly and posterior labial fusion of a 46,XX infant with 21-hydroxylase deficiency. Panel B is the growth chart of a female with classic 21-hydroxylase deficiency who experienced early puberty and early epiphyseal fusion of her bones due to excess adrenal sex steroids and obesity due to excess glucocorticoid therapy. Both likely contributed to her adult short stature. A right sided, lobulated echogenic focus measuring 2.7 x 1.0 x 1.1 cm consistent with testicular adrenal rest tissue is shown in Panel C.

Figure 4: Novel and emerging treatments in the management of congenital adrenal hyperplasia targeting various aspects of the hypothalamic-pituitary-adrenal (HPA) axis and steroid production.

Table 1- Genetic Causes and Clinical Features of the Various Forms of Congenital Adrenal Hyperplasia

Type of CAH	21-hydroxylase deficiency	11 β -hydroxylase deficiency	17 α -hydroxylase/17,20-lyase deficiency	3 β -hydroxy-steroid dehydrogenase type 2 deficiency	P450 Oxidoreductase deficiency	Lipoid adrenal hyperplasia	Cholesterol side chain cleavage enzyme deficiency
Genetic Cause (Affected Gene, OMIM#)	<i>CYP21A2</i> #201910	<i>CYP11B1</i> , #202010	<i>CYP17A1</i> , #202110	<i>HSD3B2</i> , #201810	<i>POR</i> , #201750	<i>StAR</i> , #600617	<i>CYP11A1</i> , #118485
Incidence	Classic: 1:10,000 to 1:20,000 ²² NC: 1:1,000 ²³	1:100,000 ²⁸ <u>Caucasians</u> , 1:6,000 ²⁹ Moroccan Jews* NC: unknown	1:50,000 ³⁰ Increased frequency in Brazil* ^{30,31}	Rare	Rare, 130 cases from 11 countries reported ³²	Rare, mostly Japanese, Korean, and Palestinian populations ³³	Rare, < 30 patients, mostly from Eastern Turkey ³⁴
Affected Organs	Adrenal	Adrenal	Adrenal, gonads	Adrenal, gonads	Adrenal, gonads, liver, skeletal	Adrenal, gonads	Adrenal, gonads
DSD	Classic: 46,XX NC: No	Classic: 46,XX NC: No	46,XY	Classic: 46,XY, <u>46, XX (rare)</u> NC: No	46,XX, 46,XY (variable)	46,XY NC: 46,XY (variable)	46,XY NC: 46,XY (variable)
Salt-wasting	Classic: Yes NC: No	No	No	Yes	<u>No</u>	Classic: yes NC: Minimal to none	Classic: yes NC: Minimal to none
Hypertension	No	Yes <u>NC: variable</u>	Yes	No	Yes	No	No
Postnatal Virilization	Classic: yes NC: yes	Classic: Yes NC: Yes	No	Classic: 46,XX NC: 46,XX	No	No	No
Sex Steroid	No	No	Yes	Classic: yes	Yes	Yes	Yes

Deficiency				NC: no		NC: variable	NC: variable
Other					± Skeletal malformations ± Maternal virilization		

*Due to presence of a founder mutation

CAH=congenital adrenal hyperplasia; DSD=Disorder of Sex Development; NC=nonclassic, POR= P450 Oxidoreductase; StAR= Steroidogenic acute regulatory protein.

Table 2 – Clinical Presentation and Biochemical Findings

Type/ Enzyme	Clinical Presentation	Hormonal Profile	Cosyntropin stimulation testing*	Other testing
21-hydroxylase deficiency	<p>Classic: atypical genitalia (46,XX), neonatal salt-wasting (75%), virilization < 4 year old (46,XY)</p> <p>NC: Precocious pubarche, hirsutism, <u>oligomenorrhea/amenorrhea</u>, female infertility</p>	<p>↑: 17OHP, <u>↑21-deoxycortisol</u>, androstenedione, renin</p> <p>↓: cortisol, aldosterone</p>	17OHP > 1,000 ng/dL (30 nmol/L) (several times higher for classic)	<p>NC: Early morning follicular phase 17OHP < 200 ng/dL (< 6 nmol/L)</p> <p><u>usually</u> excludes NC CAH</p>
11β-hydroxylase deficiency	<p>Classic: atypical genitalia (46,XX), virilization < 4 year old (46,XY), hypertension, hypokalemia</p> <p>NC: Precocious pubarche, hirsutism, <u>oligomenorrhea/amenorrhea</u>, female infertility, ± hypertension</p>	<p>↑: DOC, 11-deoxycortisol, androstenedione, 17OHP (mild)</p> <p>↓: Cortisol, aldosterone, corticosterone, renin</p>	11-deoxycortisol > 3 times the upper limit of normal (several times higher for classic)	
17α-hydroxylase/17,20-lyase deficiency	Adolescent female with absence of 2° sexual characteristics,	↑: DOC, corticosterone (>4000ng/dL, 115 nmol/L), progesterone	Poor response of 17OHP _{pregn} and 17OHP	

	hypertension, hypokalemia.	↓: cortisol, aldo, 17OH-pregn, 17OHP, renin DHEA, androstenedione	Elevated ratio of: DOC/cortisol, corticosterone/sex steroids	
3β-hydroxysteroid dehydrogenase type 2 deficiency	atypical genitalia (46,XX: <u>rare</u> , mild; 46,XY), neonatal salt-wasting NC: Precocious pubarche, hirsutism, <u>oligomenorrhea/amenorrhea</u> (46,XX); atypical genitalia (46,XY: mild)	↑: 17OH-pregn, DHEA, renin ↓: Cortisol, aldosterone, progesterone, 17OHP, androstenedione, DOC, 11-deoxycortisol	17OHpregn > 5000 ng/dL (150 nmol/L) Elevated Ratios pregnenolone / progesterone, 17OHpregn /17OHP	Poor testosterone response hCG stimulation in infancy
P450 Oxidoreductase deficiency	<u>Atypical</u> genitalia, ± skeletal manifestation (Antley-Bixler), ± maternal virilization	↑: pregnenolone, progesterone, 17OHP, DOC, corticosterone, ↓: DHEA, androstenedione Variable (normal or low): cortisol, aldosterone.	Variable 17OHP response, variable cortisol response (often inadequate)	Urine steroid metabolite profile shows characteristic diagnostic profile
Lipoid adrenal hyperplasia or SCC enzyme deficiency	Classic: Female genitalia, neonatal salt-wasting NC: Adrenal insufficiency (2 years- adulthood), variable gonadal function, variable genitalia (46,XY: mild)	↑: renin ↓: All steroids NC: variable	Minimal to no response NC: variable response, ↓cortisol common	Classic: Minimal response hCG stimulation Genetic testing needed to differentiate lipoid CAH and SCC

				deficiency
--	--	--	--	------------

*Administration of standard dose of 250 mcg cosyntropin ([in very low birth weight infants the dose may be reduced to 0.125mg](#)), [concomitant measurement of 17OHP, cortisol, DOC, 11-DOC, 17-OH-pregnenolone, DHEA, and androstenedione at baseline and 60 minutes distinguishes 21OH deficiency from other rarer forms of CAH.](#)

NC=nonclassic. 17OHP=17-hydroxyprogesterone. DHEA=dehydroepiandrosterone. DOC=deoxycorticosterone. 17OHpregn=17-hydroxypregnenolone. hCG=human chorionic gonadotropin. SCC=side chain cleavage.

Panel: Management of Congenital Adrenal Hyperplasia

Glucocorticoid Replacement

- Needed in classic forms of CAH, variable use in NC forms and PORD
- Children: hydrocortisone (8-15 mg/m²/day) divided into 3 doses, using the lowest dose allowing normal growth while controlling adrenal steroids
- Adolescents and adults: hydrocortisone 2-3 times daily or longer-acting^{20,67} glucocorticoids, such as prednisone (5-7.5 mg/day; 1-2 times daily), prednisolone (3-7 mg/day; 1-2 times daily) or dexamethasone (0.25-0.5 mg/day; once daily)
- Monitor for over-replacement: weight gain, central obesity, striae, stunted growth (children), declining bone mineral density
- Monitor for under-replacement: weight loss, fatigue, hyperandrogenism (21OH and 11βOH deficiency), hypertension (11βOH, 17OH deficiency and in adult POR deficiency patients)
- In women, monitor for cycle regulatory and, if appropriate, anovulation
- In males, monitor for testicular adrenal rest tissue (TART) employing testicular ultrasound from adolescence onward; if positive, then offer sperm count and motility assessment and counsel re the possibility of cryopreservation of semen

Stress Dosing

- Needed if patient receiving glucocorticoid therapy or cortisol

response to cosyntropin stimulation <18 μg/dl (500 nmol/L)

- Double or triple glucocorticoid during intercurrent illness (fever, gastrointestinal illness), surgery or trauma
- Intramuscular or subcutaneous hydrocortisone if unable to take oral glucocorticoid (home regimen). Children 50 mg/m²; adults 100 mg IV bolus followed by 200 mg over 24hours (hospital regimen)⁶⁷

Mineralocorticoid Replacement

- Needed in salt-wasting forms of CAH
- Fludrocortisone 50-300 mcg daily to achieve a plasma renin activity in the mid-normal range
- First 6-12 months of life: sodium chloride 1-2 g (17-34 mEq) daily, divided and given with feeds
- Monitor for over-replacement: hypertension, oedema, and suppressed plasma renin activity
- Monitor for under-replacement: salt-craving, orthostatic hypotension, elevated plasma renin activity
- Encourage salt intake during hot weather and conditions that promote excessive sweating. Consider seasonal adjustment of fludrocortisone dose in countries with very hot summers.

Sex Steroid Replacement

- Needed in CAH forms that result in sex steroid deficiency
- For pubertal females, oral estradiol (0.5 mg per day advanced to 1-2 mg per day); or transdermal (25 mcg per day advanced to 75-100 mcg per day) over 2-3 years; progesterone,

added following 2 years of estrogen monotherapy or when breakthrough bleeding occurs, 100-200 mg per day, or medroxyprogesterone acetate 5-10 mg per day, or norethindrone acetate 2.5-5 mg per day, for 5-10 days, in women with intact uterus³¹

- For pubertal males, intramuscular testosterone (50 mg per monthly titrated to about 200 every 2 weeks) or transdermal testosterone (titrated to 25-100g per day)³¹

Anti-hypertensive treatment

- Needed if glucocorticoid unsuccessful in treating hypertension in 11 β OH and 17OH deficiency
- spironolactone 50-200 mg per day in 1 or 2 divided doses or eplerenone 50-100 mg per day
- Calcium-channel blockers, such as amlodipine, 2.5-10 mg per day, may be used

Anti-androgen treatment

- Oral contraceptives \pm spironolactone to control hirsutism, amenorrhea in nonclassic 21OH and 11 β OH deficiency

Infertility

- Initiate glucocorticoid for nonclassic forms 21OH and 11 β OH deficiency
- Optimize glucocorticoid therapy with suppression of follicular phase progesterone (females) and

shrinkage TART (males) for 21OH and 11 β OH deficiency

- Clomiphene citrate stimulation with progesterone supplementation for hypogonadal forms CAH
- Consider In vitro fertilization (females), intra-cytoplasmic sperm injection (males)

Pregnancy

- If on glucocorticoid therapy, hydrocortisone, prednisone, or prednisolone can be used, dexamethasone should be avoided
- Increase glucocorticoid dose by 20-40%, particularly during third trimester⁶⁷
- Stress dosing for labor and delivery

Additional Monitoring Requirements

- Clinical evaluation frequently in first year of life, every 4-6 months for growing child and yearly for adults
- Patients on glucocorticoid replacement should wear emergency bracelet/card, receive sick day rule education, and carry emergency hydrocortisone kit
- Screening for psychological and sexual health issues and late-onset complications of genital surgery, if indicated
- Age-appropriate vitamin D and calcium intake and bone mineral density screening during early adulthood
- Orthopaedic management may be needed for PORD

CAH=congenital adrenal hyperplasia. NC=nonclassic. PORD= P450 Oxidoreductase deficiency.
21OH=12-hydroxylase. 11 β OH=11 β -hydroxylase. 17OH=17-hydroxylase. TART=testicular adrenal
rest tissue.

Web Appendix

[Click here to download Web Appendix: CAH Seminar_Summaries_Nov29.docx](#)