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Challenges in Sex Assignment in 46,XX Congenital Adrenal Hyperplasia due to 21-hydroxylase Deficiency and 11β-hydroxylase Deficiency in Developing Countries: Insights from an Expert Center in Indonesia

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What is already known on this topic?

Gender assignment in virilized 46,XX congenital adrenal hyperplasia (CAH) is challenging, considering many interplaying factors, such as biological function, sociocultural factors, family beliefs, and psychological outcomes. In the absence of newborn screening, many CAH individuals are late-diagnosed, leading to various practices of gender assignment across countries. The lack of knowledge regarding CAH among healthcare professionals and the absence of newborn screening added to the complexity of gender assignment of CAH individuals in Indonesia.

What this study adds?

This study presented the gender assignment practices in the only CAH center in Indonesia, highlighting the importance of timely and accurate CAH diagnosis to determine an optimal assignment for each individual. Moreover, the gender assignment approach of two types of CAH, 21-hydroxylase deficiency and 11β-hydroxylase deficiency, was presented.

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ABSTRACT

Objective: The absence of newborn screening, insufficient knowledge among medical professionals, and poor treatment adherence in congenital adrenal hyperplasia (CAH) in Indonesia causes late diagnosis. This study presents two decades of experience in gender assignment and diagnosis of 46,XX CAH.

Methods: A cohort study was carried out at a CAH referral center in Central Java, Indonesia. Data regarding clinical outcomes, molecular analysis, and sociodemographic information were taken from medical records. Participants were grouped based on current social gender, females and males. Gender at diagnosis, age at first presentation, age at first diagnosis, age at present, CAH types, virilization, puberty, birth attendant, and gender at birth decision maker significantly predict current gender identity.

Results: Among 131 individuals with 46,XX CAH, 52 (39.7%) with a sex assignment incongruent with their karyotype were included. The majority (49/52; 94.2%) had 21-hydroxylase deficiency (21OHD), while three (5.8%) had 11 beta-hydroxylase deficiency (11OHD). Individuals assigned as males at birth (3/52) had severe virilization. A change of gender occurred in 46 of 52 patients (88.46%). Midwives were the most frequent birth attendants (24/51), while pediatricians were the major decision-makers (19/51) of sex assignment.

Conclusion: In Indonesia, many 46,XX individuals with CAH were initially assigned as males due to late diagnosis, primarily caused by low awareness among healthcare professionals and exacerbated by limited medical resources and a lack of clear guidelines on sex assignment. Therefore, targeted education and standardized guidelines involving a multidisciplinary team are crucial to ensure appropriate sex assignment and care.

Keywords: Congenital adrenal hyperplasia, sex assignment, developing country, newborn screening

Introduction

Congenital adrenal hyperplasia (CAH) is a group of autosomal recessive conditions caused by enzymatic defects in steroid synthesis. Approximately 90-99% of CAH cases are caused by 21-hydroxylase deficiency (21OHD), although other causes includes 11 β-hydroxylase deficiency (11OHD), 17-hydroxylase deficiency (17OHD), *HSD3B2* deficiency, *CYP11A1* mutation, *STAR* mutation, and P450 oxidoreductase (POR) deficiency (1,2). Both 21OHD and 11OHD result in excessive adrenal androgen synthesis and cortisol deficiency leading to virilization of the external genitals in 46,XX individuals while 46,XY individuals are typically normal (3). CAH is the most frequent cause of 46,XX disorders/differences of sex development (DSD) (4).

The incidence of 21OHD (OMIM #201910) varies geographically and ethnically, with the highest rates in regions with a high degree of consanguinity. Globally, 1 in 14,000-18,000 live newborns are affected (5). 21OHD results from homozygous or compound heterozygous mutation in the *CYP21A2* genes manifesting as a clinical spectrum, from the most severe classic salt wasting (SW) type associated with cortisol and aldosterone deficiency to mild non-classic type (NCCAH), presenting with mild signs of hyperandrogenism (3). 11OHD (OMIM #202010), caused by mutations in the *CYP11B1* gene, is less common, accounting for approximately 0.2-8% of cases; hence the incidence is 1 in 100,000-200,000 live births (6,7). Similarly, 11OHD causes androgen excess, but aldosterone is unaffected. Instead, aldosterone precursors accumulate, leading to various degrees of hypertension (8).

Prenatal androgen exposure, particularly between 8-24 weeks of gestation and the early neonatal period, has long-lasting organizational effects on neurobehavioral sexual differentiation, resulting in male-typical behavior in females with 21OHD or 11OHD (9). Increased androgen also causes various degree of virilisation in females (10). The Pediatric Endocrine Society (PES) advises female assignment in 46,XX CAH newborns to maintain fertility and reproductive functionality (11). However, recent studies propose male sex of rearing for severely virilized individuals, representing an evolving perspective considering both the importance of biological functional and psychosocial factors, including family dynamics, culture, prenatal androgen exposure, and sexual function, in accordance with the Chicago Consensus (12,13).

In many Western countries, newborn screening (NBS) has enabled early CAH diagnosis and intervention to prevent life-threatening adrenal crisis and minimizing comorbidities, such as short stature, infertility, and psychological adversity (14). Nevertheless, in many developing countries, CAH diagnoses remain delayed due to the absence of NBS (15,16). Furthermore, poor awareness among many healthcare professionals leads to confusion about gender assignment. Poor infrastructure and limited transportation in Indonesia, as well as parents' fear of stigmatization and educational barriers, further delay presentation to the healthcare facilities (17).

In Indonesia, birth attendants, which may include midwife, general physician, OB/GYN, pediatrician, or traditional midwife (*paraji*), assign sex primarily based on the external genital appearance. As only "male" or "female" is legally recognized in Indonesia, ambiguous genitalia present an additional burden. Physicians often suggested giving a unisex name to avoid delay in birth registration, which must occur within 60 days. The birth certificate is vital for civil status and state recognition,

according to the Constitution of 1945. In the past, many severely virilized, late-diagnosed 46,XX CAH had to undergo a legal trial to change their gender. As a Muslim majority country, the Indonesian Ulama Council (MUI) exerts a major influence on the management of DSD individuals by issuing a *fatwa*, a religious ordinance, that does not hold constitutional power but has a significant moral impact within the Muslim society, to prohibit transsexuals from having sex reassignment surgery, but legalized gender reassignment DSD (*khuntsa*) individuals. Although the MUI's fatwa is not legally binding, it significantly shapes court decisions and medical practices (18).

Gender assignment among DSD individuals is a complex process; hence, it is obligatory to involve a multidisciplinary team consisting of at least pediatric endocrinologists, surgeons or urologists, gynecologists, geneticists, psychologists and medical ethicists. Nevertheless, not all medical centers in Indonesia have applied this approach. It has caused various sex assignments and adverse psychological outcomes, including gender dysphoria, anxiety, stress, and depression, among these individuals. This study presents two decades of experience in gender assignment and diagnosis of 46,XX CAH individuals at a single CAH referral center in a developing country, emphasizing the importance of timely and accurate diagnosis to ensure appropriate treatment in future cases.

Methods

Research Design

A retrospective cohort study was conducted on patients referred to the CAH clinic in Semarang, Central Java. The patients' data were collected from medical records between July 2004-December 2024.

Samples and Participants

All patients diagnosed by pediatric endocrinologists with CAH due to 21OHD based on clinical manifestations and 17-hydroxy progesterone (17-OHP) levels were included in this study. Patients with 11OHD CAH were diagnosed based on clinical manifestations and genetic test results. Patients with other etiologies of ambiguous genitalia were excluded.

Data Collection

The following patients' data were obtained from medical records. The diagnosis of CAH was made by an experienced pediatric endocrinologist (AU) based on: (1) clinical manifestations, including genital ambiguity, vomiting, diarrhea, and failure to thrive; (2) biochemical analyses, including levels of 17-OHP; and (3) chromosomal analysis conducted in Center for Biomedical Research (CEBIOR), Faculty of Medicine, Universitas Diponegoro, Indonesia.

The diagnosis was confirmed by mutation analysis. DNA samples were sent to the Department of Human Genetics, Radboud University Medical Centre (Radboudumc), Nijmegen, the Netherlands, for multiplex ligation-dependent probe amplification and Sanger sequencing to analyze the *CYP21A2* gene and the *CYP11B1* gene for 21OHD and 11OHD CAH, respectively. If available, genetic evaluations were considered to confirm the type of CAH and to provide genetic counselling. In 11 individuals genetic tests were not conducted, thus, electrolyte levels, showing hyponatremia and hypokalemia, were considered to differentiate between CAH types, 21OHD and 11OHD, and a history of previous hospitalization for adrenal crisis for SW CAH.

Sex Assignment

Patients presenting with a clinical suspicion of CAH, with symptoms that may have included vomiting, diarrhea, dehydration, failure to thrive, and virilization, were physically examined by a pediatric endocrinologist in our center. The degree of virilization and puberty were assessed using the Prader and Tanner stage, respectively. The patients would be referred for a biochemical investigation of electrolytes and 17OHP, to confirm CAH diagnosis. To determine the patient's karyotype, karyotyping was performed.

A psychological evaluation regarding gender assignment was not conducted for newborns, infants, and toddlers (<2 years) because the child had not developed a gender identity yet. The attending pediatric endocrinologist would explain the results and diagnosis. Based on the medical information provided by the pediatric endocrinologist, the parents made a decision regarding their child's gender.

A gender evaluation for children aged 2-7 years was conducted by a psychologist, who interviewed parents about the child's gender behavior, the child's gender preferences, and parental expectations regarding the child's gender. As gender development is dynamic and influenced by social factors, and considering the brain gender of the child, the psychologist will usually advise parents to continue observation of the child's gender behavior and preferences until adolescence, particularly concerning the possibility of the development of gender dysphoria. In most cases, girls with CAH developed masculine gender behavior and preferences, but without any confusion or dissatisfaction with the assigned gender. Children, adolescents, and adults who were late-diagnosed were referred for a psychological evaluation regarding gender assignment. A clinical psychologist and a psychiatrist conducted the assessment simultaneously and wrote a report independently to assess the individual's gender identity, gender role, and sexual orientation. Gender assignment of children aged 8 years or older was conducted using questionnaires and interviews with the patients and parents. If gender was doubted or matched with gender dysphoria according to the

DSM-5 criteria, typically triggered by the appearance of external genital or primary and secondary sexual characteristics, an interdisciplinary team meeting was held to discuss the results, outcomes, and planned management with consideration of the individual's overall well-being. A justification for gender change could not be made solely on gender behavior, such as clothing choices, playmates' preferences, and roles within a society. Instead, the individual's desire to be the opposite gender was the basis of the decision. The outcomes of the meeting, along with the consequences of choosing a certain gender, were delivered by the attending physician directly to the adult individuals and to the parents in the case of children and adolescents. Parents were given adequate time and space to make informed decisions in the best interest of their child. If a gender change was decided, it was a shared decision-making process between the team, patients, and parents.

CAH treatment was given immediately in 46,XX CAH patients assigned as females, i.e., patients with SW and simple virilizing (SV) type were given hydrocortisone (HC) combined with fludrocortisone (FC) for SW CAH and HC for SV CAH, respectively, whereas patients with 11OHD CAH were administered HC. Regular monitoring was conducted, consisting of gender satisfaction and disease evaluation control, which included signs of adrenal and SW crisis, reproductive health, and metabolic control (Figure 1).

Some of the individuals included in this study have already been reported by Ediati et al. (18) and Utari et al. (16), specifically, some who were reassigned as males and were siblings, respectively.

Ethical Considerations

All parents or caregivers provided written informed consent prior to the study, and the Diponegoro University Faculty of Medicine Health Research Ethics Committee approved the study (approval no.: 682/EC/KEPK/FK-UNDIP/XII/2024, date: 19.12.2024), guaranteeing that it complied with the 1975 Declaration of Helsinki's ethical standards.

Statistical Analysis

The categorization of the 46,XX CAH individuals were based on the current gender identity. Descriptive data are presented as frequency and percentages. Analysis was performed using SPSS, version 26.0 (IBM Corp., Armonk, NY, USA). Bivariate analysis using Fisher's exact test was conducted to determine the association between variables, including clinical characteristics and social factors. Statistical significance was defined as p value <0.05.

Results

Gender Assignment Practices

Among 131 46,XX 21OHD and 11 OHD patients managed in our center, 83 were assigned as female after birth, while 30 (22.9%) individuals received sex assignments after birth inconsistent with the karyotyping result, and 18 (13.7%) individuals were undecided (Figure 2). There were 12 individuals (23.1%) who consented to be males and refused medication after receiving information regarding the CAH condition.

Six (11.54%) were late-diagnosed and identified themselves as males who chose not to take HC or FC. Two were diagnosed with SW CAH based on genetic test results. One individual was diagnosed at the age of 3 months and received treatment until the age of 1 year. He started to be re-treated at the age of 6 years, but his treatment was irregular. At the age of 8.5 years, a re-evaluation was conducted, and he wished to remain as a male. Another individual remained untreated/chose not to be treated and was assigned as male because his parents wanted a son despite a history of adrenal crisis. The parents had been fully informed about the impact of untreated CAH, which could be life-threatening. In addition, he had precocious puberty and attained skeletal maturity at the age of ten with a final height of 135.9 cm.

Upon diagnosis of CAH, three (5.77%) individuals initially assigned as females at birth refused to take medication and chose to live as males. Their gender was reassigned at the ages of 3, 7, and 45 years. One individual was assigned and reared as female, but at age 3 years transitioned to male due to social pressure regarding the appearance of the external genitals looking more like a male's. Another individual, originally assigned as female at birth, then independently transitioned and gained real-life experience as a male, visited the center only to confirm his gender. Later, he married a woman and reported a satisfying personal life.

From a total of 18 individuals whose gender was undecided at birth, three were siblings with 11OHD who were reassigned as males. They presented with severe virilization (Prader 5) at ages 1.7, 8.5, and 2.1 years; their gender was initially undecided but re-assigned as males after the diagnosis based on their father's decision and degree of virilization. Upon diagnosis, they refused to change their gender to female. They continued to live as men, had female partners, and were employed in physically demanding jobs, including construction work and driving.

The majority (49/52; 94.23%) had pathogenic *CYP21A2* variants, of which 43 (87.76%) had SW CAH. Three individuals had died after an adrenal crisis, after multiple hospitalizations due to frequent vomiting and dehydration. Almost half (24/51; 47.1%) of the CAH patients' births were attended by midwives, followed by OB/GYN (21/51; 41.2%) and traditional midwives (6/51; 11.8%). The largest proportion of sex assignments at birth were decided by pediatricians (19/51; 37.25%). The data on birth attendants and the person in charge of the birth sex assignment of one patient were not available in the medical record.

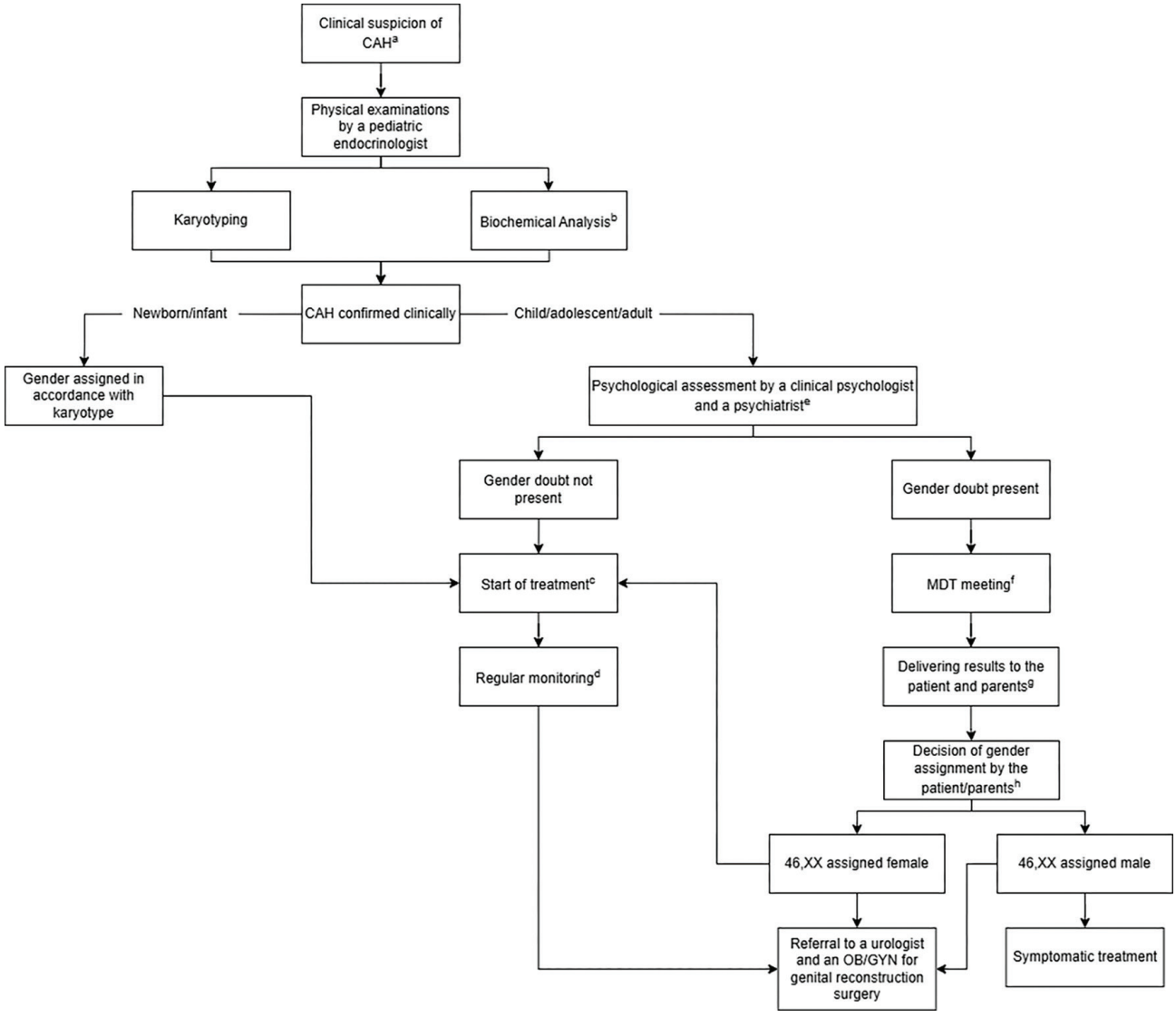

Figure 1. Flowchart of the sex assignment procedure of CAH individuals at our center

[a]Clinical suspicion of CAH includes vomiting, diarrhea, genital ambiguity, hyperpigmentation, and failure to thrive in newborns and infants. [b]Electrolytes, i.e., sodium, potassium, and chloride, and 17-OHP levels were measured. [c]Treatment comprised of hormonal, i.e., glucocorticoid and mineralocorticoid, and symptomatic, i.e., antihypertensive for 11OHD and NaCl for CAH SW. [d]Symptoms of adrenal insufficiency, menstrual cycle, libido and erection, sexual health, pubertal development, height, body mass index, blood pressure, bone age, and scrotal or ovarian ultrasound were monitored as indicated. [e]Psychological assessment of the individual's gender identity, gender role, and sexual orientation was conducted by a clinical psychologist and a psychiatrist. Reports were made independently. [f]A multidisciplinary team consisted of a pediatric endocrinologist, a geneticist, a clinical psychologist, a psychiatrist, a urologist, an OB/GYN, and a genetic counsellor. [g]Results were delivered to the patients and parents of children or adolescents. For adults, the results were received solely by them. [h]The gender decision was made by the parents considering the child's well-being. Adults with CAH made their own decision. 11OHD, 11β-hydroxylase deficiency; 17-OHP, 17-hydroxyprogesteron; CAH, congenital adrenal hyperplasia; MDT, multidisciplinary team.

Predictors of Gender at Present

Table 1 shows a significant difference in age at diagnosis between females and males with 46,XX CAH, in which 87.5% of females (n=40) were diagnosed at age <1 year, compared to 8.3% in the male group (n=12) (p<0.001). In contrast to 62.5% females presenting with Prader 3, severely virilized individuals were significantly more likely to have male gender identity (p<0.008), as evidenced by 41.7% and 25.0% males presenting with Prader 4 and 5, respectively. The involvement of healthcare professionals at the birth of CAH individuals significantly predicted their current gender (p<0.001), in which all females were attended by either a midwife or OB/GYN, and 54.5% of males were attended by non-healthcare professionals (i.e., traditional midwives). Due to the limited sample size, sensitivity analysis of the present gender subgroups was not conducted.

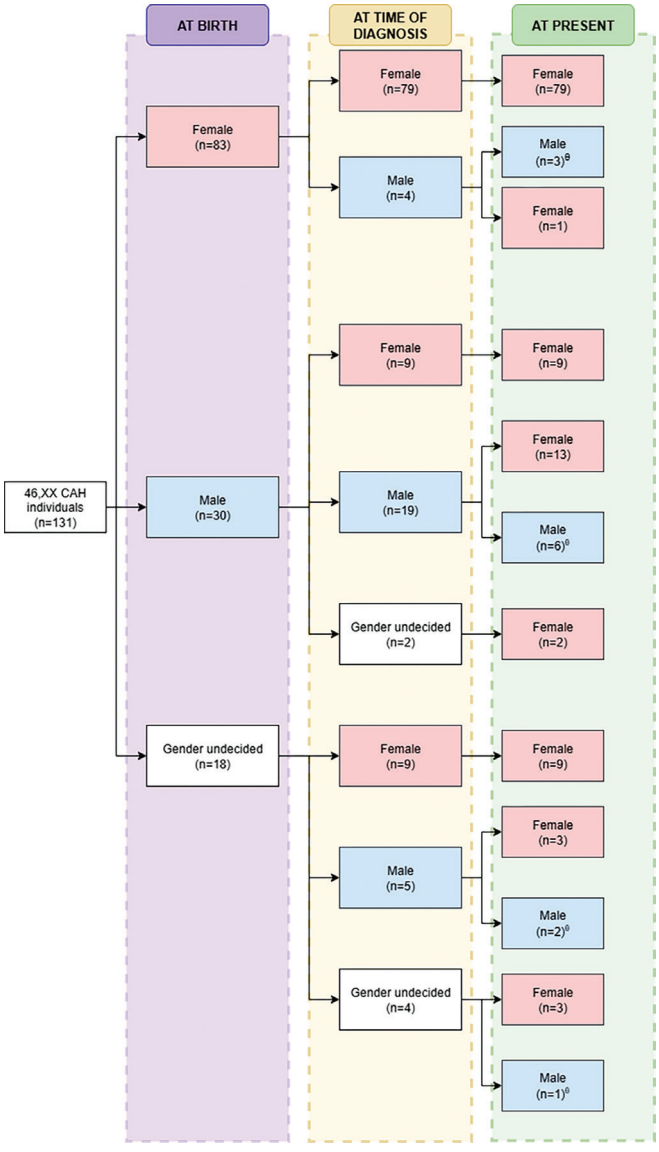

Figure 2. Gender assignment odyssey of 46,XX CAH individuals in Indonesia

θCAH individuals who were reassigned as males gave informed consents not to take medication, i.e., HC and FC, due to their wish to remain as males.

Discussion

Various gender assignment practices in CAH individuals remain common in Indonesia, even after two decades of experience. The majority of 46,XX CAH individuals in this study were late-diagnosed, which is comparable to a previous study in the context of lack of NBS for CAH (19). In Indonesia, the pilot project of CAH NBS only started in 2024 and expanded nationally at the beginning of 2025. Individuals who received earlier recognition were mostly attended by healthcare professionals, midwives and OB/GYN, and depicted an increasing tendency to be assigned as females. According to the Decree of the Minister of Health, uncomplicated spontaneous birth was prioritized to be assisted in primary healthcare facilities, such as sub-district health centers (*Puskesmas*), midwife private practices, and primary doctor clinics, attended by midwives or general physicians (20). Although the Indonesian government increased the number of midwives at the community level and healthcare facilities in both urban and rural areas, only 55.2% of mothers sought help with childbirth from healthcare professionals, such as general physicians, midwives, hospitals, maternity homes, or other health centers (21). This indicates a significant reliance on non-medical professionals, including traditional midwives (*paraji*) who lack formal medical training.

The majority of 46,XX CAH individuals in Indonesia had the most severe SW type. Children with CAH were usually born with an uncomplicated pregnancy and spontaneous labor, hence, most children born in primary and secondary healthcare facilities were not immediately referred to the central hospital to receive an appropriate diagnosis and treatment. Referrals typically occurred after frequent episodes of vomiting and dehydration without improvement after receiving treatment, which are the characteristics of the most severe SW type (16,22,23). Delayed referral is a common challenge found in developing countries, as reported from Bangladesh, Sri Lanka, and Malaysia (24,25,26). Many Indonesian healthcare professionals lack knowledge about CAH, including the atypical presentation of external genital, clinical manifestation of adrenal insufficiency, genetic etiology, laboratory workup, and management (27). For example, genetics was only included in the list of midwives' standard competencies in 2020 (28).

Some 46,XX individuals with CAH were initially treated, but due to a lack of compliance, their conditions were poorly controlled. Upon monitoring, they were offered continuation of medication, but, despite understanding the impending adrenal crisis, chose not to take medication which may lower androgen levels, because they would like to remain as males. Our data showed that adherence to treatment remained an issue, despite the increased availability of CAH medications (29). CAH individuals in a developing country had to travel considerable distances and a spend a substantial amount of time to receive care at the tertiary hospital (30). The need for lifelong medications might reduce individuals' motivation to adhere to treatment. Adding to the complexity were inadequate information and myths about the side effects of taking lifelong medications, which are commonly believed in developing countries (31,32). Conversely, a previous study in Sweden reported good treatment adherence in children and adults with CAH, with better adherence observed in SW, the most severe CAH (33).

Table 1. Sex assignment practices in 46,XX CAH individuals

	Current gender		p value
	Female n (%)	Male n (%)	
Sex at birth (n=52)			0.064
Male	24 (60.0)	6 (50.0)	
Female	1 (2.5)	3 (25.0)	
Undecided	15 (37.5)	3 (25.0)	
Gender at diagnosis (n=52)			**0.004***
Male	17 (42.5)	11 (91.7)	
Female	18 (45.0)	1 (8.3)	
Undecided	5 (12.5)	0 (0.0)	
Age at first presentation (n=52)			**<0.001***
<1 y	37 (92.5)	0 (0.0)	
1-5 y	2 (5.0)	3 (25.0)	
5-11 y	1 (2.5)	5 (41.7)	
11-18 y	0 (0.0)	2 (16.7)	
>18 y	0 (0.0)	2 (16.7)	
Age at first diagnosis (n=52)			**<0.001***
<1 y	35 (87.5)	1 (8.3)	
1-5 y	4 (10.0)	3 (25.0)	
5-11 y	1 (2.5)	4 (33.3)	
11-18 y	0 (0.0)	2 (16.7)	
>18 y	0 (0.0)	2 (16.7)	
Age at present (n=46)			**<0.001***
<1 y	33 (82.5)	0 (0.0)	
1-5 y	6 (15.0)	2 (33.3)	
5-11 y	1 (2.5)	3 (50.0)	
11-18 y	0 (0.0)	0 (0.0)	
>18 y	0 (0.0)	1 (16.7)	
CAH types (n=52)			**<0.001***
SW	39 (97.5)	4 (33.3)	
SV	1 (2.5)	5 (41.7)	
11OHD	0 (0.0)	3 (25.0)	
Degree of virilization (n=52)			**0.008***
Prader 1	0 (0.0)	0 (0.0)	
Prader 2	2 (5.0)	1 (8.3)	
Prader 3	25 (62.5)	3 (25.0)	
Prader 4	13 (32.5)	5 (41.7)	
Prader 5	0 (0.0)	3 (25.0)	
Puberty at diagnosis (n=52)			**<0.001***
Yes	0 (0.0)	9 (75.0)	
No	40 (100.0)	3 (25.0)	

Table 1. Continued

	Current gender		p value
	Female n (%)	Male n (%)	
Treatment status (n=52)			**<0.001***
Treated (hormonal treatment)	36 (90.0)	0 (0.0)	
Untreated	0 (0.0)	10 (83.3)	
Loss to follow-up	1 (2.5)	2 (16.7)	
Died	3 (7.5)	0 (0.0)	
Birth attendant (n=51)†			**<0.001***
Midwife	20 (50.0)	4 (36.4)	
OB/GYN	20 (50.0)	1 (9.1)	
Traditional midwife (*paraji*)	0 (0.0)	6 (54.5)	
Decision maker of gender at birth (n=51)††			**0.011***
Midwife	12 (30.0)	4 (36.4)	
OB/GYN	2 (5.0)	0 (0.0)	
Pediatrician	18 (45.0)	1 (9.1)	
General physician	1 (2.5)	0 (0.0)	
Traditional midwife (*paraji*)	0 (0.0)	3 (27.3)	
Parents	7 (17.5)	3 (27.3)	
Genetic variants (n=82)§			
R356W	53.1	13.6	
I2G	28.1	31.8	
Exon 1-7 del	6.3	0.0	
P30L	6.3	0.0	
W22X	3.1	0.0	
p.Trp406*	3.1	0.0	
p.Trp20*	0.0	4.5	
I172N	0.0	4.5	
Exon 1-3 del	0.0	4.5	
p.Ile386del	0.0	4.5	
p.Gln196*	0.0	4.5	
R356W/Microconversion in the promoter region (c.-126T; c.-113G>A; c.-110T>C; c.-103A>G) [~80% less active transcript]	0.0	4.5	
p.Val252fs (CYP11B1)	0.0	27.3	

*Statistically significant ($p<0.05$)
@There were 6 individuals who were assigned male at birth and stayed as males.
#Puberty was determined by pubarche (Tanner P2).
†Data were unavailable from 1 male patient due to loss to follow-up.
††Data were unavailable from 1 male patient due to loss to follow-up.
§Allele frequency was calculated from the genetic test results of 41 individuals. Allele frequency was presented in percentage. No comparative analysis for the allele frequency variable was conducted.
¶Individuals were siblings. Their father decided their sex based on the male-like appearance of their external genitalia.
N/A: not available; 11OHD: 11 β -hydroxylase deficiency; CAH: congenital adrenal hyperplasia; SV: simple virilizing; SW: salt wasting.

Individuals with CAH in developing countries missed receiving timely diagnosis and treatment. The lack of laboratory analysis facilities in Indonesia, especially outside of Java, hindered diagnosis in our cohort. With prolonged time to diagnosis, CAH individuals were exposed to extended periods of elevated androgen levels which affected permanent neural organizational changes in the brain and, as a consequence, caused male-typical behaviors and toy choices in females with CAH (34). The masculinization effect from the androgen persisted and played a role in the establishment of male gender identity in 46,XX CAH (35). Therefore, after thorough evaluation with a clinical psychologist, individuals diagnosed in childhood or beyond determined to remain males owing to the androgen masculinizing effect on their gender behavior and identity. This finding was in accordance with a prior study that illustrated a mid-childhood conversion to male in late-diagnosed 46,XX CAH individuals in Bangladesh (36). In contrast, transition to male in 46,XX individuals with CAH has rarely been reported in developed countries due to early diagnosis and adequate access to medication. Nevertheless, this presented a complex situation because assigning a 46,XX CAH individual as male could deprive them of the opportunity of having children; the reproductive function would be preserved in 46,XX CAH females who were treated promptly and appropriately (3).

Gender identity issues and distress were commonly observed in DSD individuals, which led to gender dysphoria (37). Late-identified DSD individuals have reported experiencing more emotional and behavioral issues, including social isolation, particularly in adult women (38). Compared to children with DSD who were raised as boys, those who were raised as girls showed a high level of gender confusion. The same study also reported that women with DSD who received no treatment exhibited behavior inconsistent with their gender roles and were presumably more dissatisfied with their gender identity (18). Findings from some Sri Lankan and Indian 46,XX CAH populations revealed that a subset of individuals raised as females had gender dysphoria (25,39). Nonetheless, a meta-analysis found that the prevalence of gender identity issues was higher in CAH-raised males compared to females (40).

The majority of 46,XX CAH individuals reassigned as female were diagnosed before the age of one year. At this age, the primary social interactions were within the family where parents had the strongest impact on gender role development in their children (41). Later, children start forming stereotypes for both sexes at the age of two to three years, where they begin socializing beyond the family (42). Social stereotypes and pressure may influence the child's emerging gender identity, as seen in one of the individual within the "assigned male and remained as male" group.

However, a study by Dessens (43) revealed that some previously misassigned females with CAH at birth had difficulty with society's acceptance of gender reassignment. This was aggravated by preconceived notions and fear of social rejection. These stereotypes of how someone should behave to conform to a certain gender increased the burden on females with CAH, particularly if they were late-diagnosed and priorly assigned as males. In some cases, societal values further complicate the decision of gender assignment. In our society that prefers male children over females, some parents still choose to raise their 46,XX child as a male, even after diagnosis. Having a son was considered superior to having a daughter because, although a male is infertile, he can still hold a high place in a community and get a better job compared to a female. This was a common practice in Indonesia as patriarchal beliefs were still held by many, especially in rural areas (44).

Most 46,XX CAH in our cohort came with a moderate to severe degree of virilization (Prader 3-5), and more than half were assigned as male at birth. The appearance of external genitalia had a great impact on the gender assignment process, in which Prader 1-3 were more likely to be assigned females, but Prader 4-5 were apt to be assigned males, particularly due to the complexity of feminizing surgery, which may include worse outcomes, more complications, and greater assessment challenges (45). Both DSD and controls in a study by Chowdhury et al. exhibited a more masculine identity when they had more severely virilized external genitalia (46).

The development of one's gender is a fluid process with various factors interplaying, including potential for future fertility, hormonal therapy, feminizing or masculinizing surgery, psychological implications, and sociocultural factors (3). The latter were very diverse between populations, hence an individualized and measured approach needs to be considered (15,24,25,44). In the CAH Clinical Guidelines, an infant born with 46,XX CAH should be assigned female. This recommendation was made reflecting the condition in developed countries where early diagnosis was possible due to the availability of NBS (5). Previous studies reported that male assignment in 46,XX CAH could be an option in severely masculinized individuals, as they showed a good quality of life (QoL) (12). An emphasis on social and cultural preferences needed to be considered in assigning a person's gender, for example in Middle and Far Eastern countries, where males were considered to have a higher status than females (47). According to the Clinical Guidelines for the Management of Disorders of Sex Development in Childhood, the management of a person with a DSD condition must be individually tailored. A patient-centered care approach from a multidisciplinary team should consider the uniqueness of each person; hence, the best decision needs to be made in the patient's best interest (48). A psychologist or a psychiatrist trained in DSD care played an

integral part in gender assignment or reassignment, decision regarding surgery or hormonal treatment, and assessment of short or long-term outcomes. However, the DSD multidisciplinary team was only available in Semarang, Indonesia, bringing challenges to the provision of holistic and comprehensive care for the patients and their families (49).

Due to its potential to be life-threatening, CAH must be ruled out in every child born with genital ambiguity, and the attendants should contact healthcare professionals familiar with the disease. Although genital ambiguity is present only in 46,XX CAH, male gender assignment was common in a region without NBS (5). Several studies reported that only a small portion of severely virilized 46,XX CAH raised females reported dissatisfaction with their gender. Stigmatization related to cross-gender behavior and atypical physical appearance has been identified as a stress predictor in both adults with DSD and parents of children with DSD (23). In India, DSD individuals were labelled as "*hijra*" and received discrimination, which resulted in lower QoL (50). In developing countries, fertility and future marriage remained a concern for parents of female CAH (30).

A guideline for sex assignment in CAH individuals in Indonesia has not been established. The beginning of CAH NBS in Indonesia in 2025 represents a significant advance, offering a promising opportunity for early identification and treatment for affected individuals, thereby anticipating serious outcomes such as adrenal crisis, delayed sex assignment, and gender dysphoria. Nevertheless, national CAH guidelines for sex assignment need to be established. Furthermore, the development of effective education strategies for healthcare professionals will improve the knowledge and skills of healthcare professionals regarding CAH. Thus, its implementation should be made at primary, secondary, and tertiary care levels. Future research on the long-term evaluation of the psychological and social impact of late diagnosis should be conducted in resource-limited settings.

Study Limitations

This was a descriptive study in a single national DSD referral center, but it can provide useful insights into the past and changing practice concerning individuals with CAH. This study relied on retrospective data, which may be affected by incomplete medical records. Moreover, findings from a single referral center may not fully represent the sex assignment practices of CAH individuals in Indonesia. This study was not able to report the impact of sex assignment on the CAH individuals. Hence, future studies are needed to provide broader insights into CAH sex assignment practices in developing countries.

Conclusion

Various sex assignment practices affecting 46,XX CAH individuals in Indonesia remain, caused by low awareness among healthcare professionals and a lack of laboratory facilities, leading to late diagnosis. Gender at diagnosis, age at first presentation, at first diagnosis, and at gender reassignment, types of CAH, degree of virilization, puberty status, state of treatment, birth attendant, and decision maker for gender at birth were significant predictors of gender at present. Treatment compliance issues exposed these individuals to excessive androgen levels, causing male-gender behavior. Cultural, religious, and family values played an important role in shaping gender identity. In Indonesia, development of national CAH sex assignment guidelines and targeted education for healthcare professionals is necessary to ensure better long-term outcomes for individuals affected by CAH.

Ethics

Ethics Committee Approval: Diponegoro University Faculty of Medicine Health Research Ethics Committee approved the study (approval no.: 682/EC/KEPK/FK-UNDIP/XII/2024, date: 19.12.2024).

Informed Consent: All parents or caregivers provided written informed consent prior to the study.

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Footnotes

Authorship Contributions

Concept: Irene Astrid Larasati, Agustini Utari, Tri Indah Winarni, Design: Irene Astrid Larasati, Agustini Utari, Tri Indah Winarni, Data Collection or Processing: Irene Astrid Larasati, Agustini Utari, Annastasia Ediati, Analysis or Interpretation: Irene Astrid Larasati, Agustini Utari, Annastasia Ediati, Hedi L. Claahsen - van der Grinten, Tri Indah Winarni, Literature Search: Irene Astrid Larasati, Agustini Utari, Writing: Irene Astrid Larasati, Agustini Utari, Annastasia Ediati, Hedi L. Claahsen - van der Grinten, Tri Indah Winarni.

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