Fertility in Women with Nonclassical Congenital Adrenal Hyperplasia due to 21-Hydroxylase Deficiency

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Objective: In contrast to subfertility often reported in women suffering from the classical form of congenital adrenal hyperplasia (CAH) due to 21-hydroxylase deficiency, fertility in nonclassical CAH (NC-CAH) has been rarely studied. Our objective was to evaluate fertility in NC-CAH women.

Material and Methods: We studied 190 NC-CAH women (161 probands + 29 first degree relatives). Only 20 probands had consulted for infertility (12%), either alone or associated with hirsutism or menstrual cycle disorders. The diagnosis was established on post-ACTH 17-hydroxyprogesterone 10 ng/ml or greater and further characterized by *CYP21A2* gene analysis.

Results: Ninety-five of the 190 women wanted pregnancy (aged 26.7 ± 8.9 yr); 187 pregnancies occurred in 85 women, which resulted in 141 births in 82 of them. Ninety-nine pregnancies (52.9%) occurred before the diagnosis of NC-CAH (96 spontaneously and three with ovulation inducers) whereas 98 occurred after diagnosis (11 spontaneously and 77 with hydrocortisone treatment); 83% of pregnancies were obtained within 1 yr. The rate of miscarriages was 6.5% for pregnancies obtained with glucocorticoid treatment *vs.* 26.3% without. Two of the 141 infants (1.5%) were born with classical CAH.

Conclusion: Subfertility is mild in NC-CAH. However, the rate of miscarriages is lower in pregnancies occurring with glucocorticoid treatment and argues for treating NC-CAH women wanting pregnancy. In addition, considering the high rate of heterozygotes for *CYP21A2* mutations in the general population, it is essential to genotype the partner of patients with a severe mutation to predict the risk of classical CAH and offer genetic counseling. **(*J Clin Endocrinol Metab* 95: 1182–1190, 2010)**

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Abbreviations: A, Δ4-Androstenedione; CAH, congenital adrenal hyperplasia; 21dF, 21-deoxycortisol; NC-CAH, nonclassical CAH; 21OH, 21-hydroxylase; 17OHP, 17-hydroxyprogesterone; T, testosterone; TTP, time to pregnancy.

Congenital adrenal hyperplasia (CAH) due to steroid 21-hydroxylase (21OH) deficiency is one of the most common inborn endocrine disorders and is inherited as an autosomal recessive disease (1). Molecular abnormalities of the *CYP21A2* gene coding for the steroid 21OH enzyme lead to various degrees of impaired cortisol and aldosterone synthesis and androgen excess. There is marked polymorphism of clinical expression, depending on severity of the enzymatic defect. Besides the classical form (CAH), due to complete or severe 21OH deficiency generally revealed at birth, there exists the nonclassical form (NC-CAH), due to partial enzymatic defect, with late-onset symptoms and diagnosis in childhood or after puberty (2–4).

Subfertility, a reduced capacity to conceive, has often been reported in women with the classical form of CAH (5–12) and may be due to different mechanisms such as genital abnormalities or anovulation. However, there are few data concerning fertility in women with NC-CAH (13–15), and menstrual irregularities or subfertility have been only incidentally reported.

The phenotype of NC-CAH patients is classically determined by the less severe *CYP21A2* mutation with the highest residual 21OH enzymatic activity. Nevertheless, several studies have reported that 27–76% of NC-CAH patients are carriers of a severe mutation (16–18). These patients may give birth to a child with the classical form of CAH if their partner is also carrying a severe mutation.

It therefore appeared interesting to evaluate retrospectively fertility in 190 women investigated and followed up in our department for NC-CAH (17) and also their risk of giving birth to a child with the classical form of CAH.

Patients and Methods

Patients

NC-CAH due to partial 21OH deficiency was diagnosed in 190 women, including 161 unrelated probands who had consulted in our department and 29 affected first-degree female relatives. Only 20 probands had consulted for infertility (12%), either alone or associated with other symptoms. Most probands had consulted for hirsutism and/or menstrual cycle disorders, which were present in 78 and 61%, respectively. (Table 1). They had consulted at the age of 23.4 ± 8.8 yr (mean ± SD, range 13–52 yr). NC-CAH due to 21-OH deficiency was diagnosed on 17-hydroxyprogesterone (17OHP) plasma level above 10 ng/ml after ACTH stimulation and was further characterized by molecular analysis of the *CYP21A2* gene in 124 probands.

The investigation of the probands' family members led to the identification of 29 female relatives with the same biological profile as probands, who were therefore diagnosed as NC-CAH patients: 16 sisters, six mothers, and seven daughters. These females had not spontaneously consulted, but an agreement was obtained for clinical and hormonal investigations. They were 26.2 ± 16.3 yr old (range 1–52 yr) at the time of diagnosis. Seventeen were postpubertal and premenopausal. Hirsutism was observed in five (17%) and subfertility with oligomenorrhea in one. These relatives were considered to have the same 21OH deficiency as the index case of their family based on post-ACTH 17OHP level higher than 10 ng/ml and two different methods, depending on the period of investigation of the patients and their relatives: HLA typing up to 1986 and then molecular analysis of the *CYP21A2* gene.

Hormone determinations

The patients underwent routine etiological investigations for hirsutism, menstrual cycle disorders, and/or infertility, and the following hormones were assayed: basal plasma estradiol, testosterone (T), Δ4-androstenedione (A), 17OHP, and 21-deoxycortisol (21dF), measured before and 1 h after im administration of 0.250 mg synthetic ACTH [ACTH-(1-24), Synacthen; Ciba SA, Gron, France]. Biological evaluation of gonadotropin function included measurements of plasma LH and FSH before and after iv injection of 100 μg GnRH. Hormone levels were measured during the follicular phase of the menstrual cycle. Blood samples were collected at 0800 h under basal conditions. Plasma hormones were measured bu RIA as previously described (18–21).

Molecular analysis

Molecular analysis of *CYP21A2* was performed in 144 female patients, including 124 unrelated probands and 20 family members (16, 17). A blood sample was collected from each patient after obtaining written informed consent. Molecular analyses were performed as recently reported (17).

Fertility evaluation in NC-CAH patients

Among the 190 NC-CAH female patients of this single-center study, 90 are still regularly followed up in our department and could be interviewed during their visits. The others were mailed a questionnaire or were interviewed by phone. If necessary, data not provided by the patient were obtained from the patient's practitioners. The main data collected concerned menstrual cycle existence and length; hormone assays; pelvic ultrasonography; pregnancy desire; number of pregnancies obtained; spontaneously or with treatment (glucocorticoid or ovulation inducers); time to pregnancy (time span exposed to unprotected intercourse until conception); outcome (birth, miscarriage, or voluntary abortion); term of birth or miscarriage; singleton or multiple pregnancy; and complication events. Other factors of infertility were also recorded.

The time span or number of menstrual cycles exposed to unprotected intercourse until conception is the time to pregnancy (TTP). For pregnancies that resulted in voluntary abortion, TTP was considered to be undefined (22).

Statistical analysis

Descriptive statistics used numbers and percentages for qualitative variables and means, SDs, medians, and ranges for quantitative variables. Comparisons between groups were tested using the χ^2 or Fisher's exact test for qualitative variables and two-sample Wilcoxon's test for quantitative variables. Test choices were based on group size. A multivariate analysis of risk factors for spontaneous miscarriage was performed using stepwise logistic regression. Potential factors included in the regression were variables with $P < 10\%$ and missing data rates less

TABLE 1. Clinical and hormonal characteristics of the 190 NC-CAH female patients

	Total patients (n = 190)	Probands (n = 161)	Relatives (n = 29)	Patients wanting pregnancy (n = 95)
Clinical characteristics				
Age at diagnosis (yr)	23.6 ± 9.7	23.4 ± 8.8	26.2 ± 16.3	26.7 ± 8.9
Age at menarche (yr)	12.5 ± 1.6	12.8 ± 0.3	12.3 ± 1	12.5 ± 1.5
Premature pubarche	10%	11.3%	0.3%	4%
BMI (kg/m^2)	24 ± 4.6	23.8 ± 4.5	25.7 ± 7.8[a]	24.3 ± 4.6
Height (cm)	162 ± 6	161 ± 4	165 ± 1[a]	160 ± 6
Hirsutism	71.8%	78%	17%	76%
Primary amenorrhea	8.2%	8.9%	0	9.3%
Secondary amenorrhea	4.4%	4.7%	0	3.5%
Oligomenorrhea	46.2%	48%	21%	43.7%
Regular menstrual cycles	41.2%	38%	78.5%	44%
Infertility	11%	12%	0.3%	22%
Hormones values, mean ± SD (range)				
Basal 17OHP (ng/ml)	11.5 ± 13.4 (0.59–67.0)	13.2 ± 16.4 (0.6–67.0)	7.9 ± 10.1 (1.00–41.2)	11.3 ± 11.6 (1.2–67.0)
Post-ACTH 17OHP (ng/ml)	40.3 ± 20.5 (10–108)	43.34 ± 29.01 (10–108)	36.3 ± 16.9 (11.7–70.)	45.1 ± 24.0 (11–108)
Basal 21dF (ng/ml)	2.7 ± 4.1 (0.04–20.6)	3.0 ± 4.3 (0.19–20.6)	1.6 ± 2.8 (0.04–9.5)	1.8 ± 1.7 (0.19–6.05)
Post-ACTH 21dF (ng/ml)	13.4 ± 14.5 (1.2–103)	14.43 ± 15.8 (1.2–103)	9.4 ± 5.6 (3.9–19.4)	11.2 ± 7.5 (1.2–24.8)
T (ng/ml)	0.84 ± 0.49 (0.2–2.9)	0.86 ± 0.50 (0.2–2.9)	0.50 ± 0.30 (0.2–1.8)[b]	0.89 ± 0.53 (0.2–2.9)
A (ng/ml)	4.24 ± 2.57 (0.98–16.50)	4.42 ± 2.60 (0.98–16.50)	2.30 ± 1.00 (1.50–4.32)[b]	4.11 ± 2.7 (1.20–16.50)
Basal FSH (mIU/ml)	4.5 ± 1.9 (1–9)	4.6 ± 2.0 (1–12)	6.6–1.4[c]	4.4 ± 1.6 (1.0–8.0)
Post-GnRH FSH (mIU/ml)	12.0 ± 9.0 (2.6–32)	12.2 ± 9.0 (2.6–32)	3.1	12.7±11.8 (3.0–32)
Basal LH (mIU/ml)	5.3 ± 4.4 (0.37–15)	5.4 ± 4.5 (0.37–15)	2.3–0.7[c]	5.3 ± 4.16 (0.8–15)
Post-GnRH LH (mIU/ml)	38.0 ± 30.0 (6.5–86)	38.3±30.0 (6.5–86)	15	38.7 ± 32.1 (6.5–86)

BMI, Body mass index.

[a] For pospubertal patients; [b] for 17 pospubertal and premenopausal patients; [c] available for only two female relatives.

than 20%. The final model retained variables with $P < 5\%$ using the Wald test. Two cumulative incidence curves of TTP were estimated: the first for the entire sample of women wanting pregnancy and the second conditioned by the pregnancy occurrence. All tests were two tailed, with a threshold P value of 5%. Computations were performed using the SAS version 8 statistical package (SAS Institute, Cary, NC).

Results

Characteristics of the 161 probands

Their clinical phenotype and hormonal characteristics are presented in Table 1. Age at menarche was 12.8 ± 0.3 yr (mean ± SD, range 9–17 yr). Mean plasma basal 17OHP level was 13.2 ± 16.4 ng/ml and rose to 43.3 ± 29 ng/ml after ACTH stimulation. Mean plasma T and A levels were 0.86 ± 0.50 and 4.4 ± 2.6 ng/ml, respectively. Basal plasma FSH and LH were 4.6 ± 2.0 and 5.4 ± 4.5 mIU/ml and rose to 12.2 ± 9.0 and 38.3 ± 30.0 mIU/ml, respectively, under GnRH test. In 53 patients, pelvic ultrasonography had been performed in the context of menstrual cycle disorders or consultation for infertility; in 27 (51%) of these 53 patients, multiple ovarian follicles were observed suggesting polycystic ovaries.

Molecular analysis of the *CYP21A2* gene was performed and previously reported in 124 probands (18). Briefly, 108 patients (85.7%) had the mild V281L mutation, and 31 of these (24.6%) were homozygous for this mutation. It should be underlined that 79 (62.5%) had at least one severe mutation and surprisingly four had two severe mutations.

Characteristics of the 29 affected female relatives

Seventeen of the affected female relatives were postpubertal, and age at menarche was 12.3 ± 1.14 yr (range 10–14 yr). For the 29 female relatives, mean basal plasma 17OHP levels was 7.9 ± 10.1 ng/ml and increased to 36.3 ± 16.9 ng/ml after ACTH stimulation. Mean basal plasma 21dF was 1.6 ± 2.8 ng/ml; it rose to 9.4 ± 5.6 ng/ml after ACTH. Plasma T and A levels were 0.50 ± 0.30 and 2.3 ± 1.0 ng/ml, respectively, for the whole group of female relatives. These levels were 0.60 ± 0.30 and 2.4 ± 1.0 ng/ml, respectively, for the 17 postpubertal ones.

Molecular analysis of the *CYP21A2* gene was performed in 20 of these 29 female relatives; 17 (85%) had the V28IL mutation, of whom three were homozygous for this mutation; 13 (65%) had one severe mutation.

Because these female patients had the same NC-CAH disease as the probands, they were added to the 161-patient cohort for the study of fertility. The clinical and hormonal characteristics of the entire cohort of 190 NC-CAH women are shown in Table 1.

Occurrence of pregnancies and their outcomes in NC-CAH women wanting pregnancy

Among the 190 female NC-CAH patients, we obtained information from 168 (88.4%); 95 wanted pregnancy (85 probands and 10 family members) (Table 1); 85 conceived and 82 (86.3%) gave birth. One hundred eighty-seven pregnancies occurred, which resulted in 141 live births (75.4%) including 135 singletons and three sets of twins. The other outcomes were two ectopic pregnancies, one therapeutic abortion for toxoplasma gondii infection, 12 voluntary abortions, and 34 early spontaneous fetal miscarriages between 6 and 12 wk of amenorrhea. These miscarriages accounted for 19.4% of pregnancies when voluntary abortions were excluded. No delivery occurred before 33 wk of amenorrhea. Five of the 135 singleton deliveries (3.7%) occurred between 33 and 37 wk of amenorrhea. The terms of the three twin births were 36, 37, and 38 wk, which makes a total rate of 4.3% moderate preterm deliveries.

Finally, 1.97 pregnancy and 1.48 live births occurred per woman wanting pregnancy. The mean age of patients was 27.9 ± 4.4 yr at the time of their first pregnancy and 29.8 ± 5.1 yr for all pregnancies.

Pregnancy occurrence and outcome according to the time of NC-CAH diagnosis and treatments

Ninety-nine pregnancies occurred before the diagnosis of NC-CAH (52.9%), 96 spontaneously, and three after induction of ovulation (one with clomiphene citrate and two with gonadotropin treatment). Eighty-eight pregnancies occurred after the diagnosis of NC-CAH, 11 spontaneously, and 77 with glucocorticoid treatment.

When all pregnancies are considered as a whole, 107 (57%) were spontaneous, 96 before and 11 after NC-CAH diagnosis (Table 2). The outcomes of these spontaneous pregnancies were 67 live births (62.6%), 27 miscarriages (25.2%), two ectopic pregnancies (1.8%), one therapeutic abortion, and 10 voluntary abortions (9.3%). Seventy-seven pregnancies (41%) occurred with glucocorticoid treatment, administered alone (n = 62) or associated with ovulation inducers (n = 15), principally clomiphene citrate. Glucocorticoid treatment was hydrocortisone (17.7 ± 3.7 mg/d) for 52 pregnancies and dexamethasone (0.25 to 0.50 mg/d) for 10. These pregnancies resulted in 73 births (92.5%), including three sets of twins, five miscarriages (6.5%), and two voluntary abortions (2.5%). No difference in the term of deliveries was observed with or without glucocorticoid treatment. One patient experienced acute adrenal insufficiency due to repeated first-trimester pregnancy vomiting. The patient recovered and the pregnancy went on after adjustment of hydrocortisone treatment.

TABLE 2. Pregnancy occurrence and outcome according to glucocorticoid treatment (Tt)

	Pregnancies without corticoid Tt		Pregnancies with corticoid Tt	
	Spontaneous n	With ovulation inducers n	With glucocorticoid alone n	With glucocorticoid + ovulation inducers n
Total pregnancies	107	3	62	15
Births	67	1	57	16
Abortions and EP	13	0	2	0
Miscarriages	27 (25.2%)	2 (26.3%)	3 (4.8%)	2 (6.5%)

EP, Ectopic pregnancy.

The characteristics of the patients who obtained pregnancy without or with glucocorticoid treatment are shown in Table 3. The patients who obtained pregnancy with glucocorticoid treatment were significantly younger at the time of diagnosis, with significantly more frequent hirsutism and higher plasma T and A levels ($P < 0.05$). In addition, they more frequently presented menstrual cycle disorders. In both groups, the distribution of *CYP21A2* mutations was mild/mild for one of three and mild/severe for two of three, as in the whole cohort (17).

Among the our patients with two severe mutations, three patients wanted pregnancy; the fourth is 17 yr old. Two patients obtained two pregnancies each; they occurred before the diagnosis of NC-CAH without any treatment and resulted in two births for one and one birth and one early miscarriage for the other. The third patient obtained three pregnancies with hydrocortisone treatment after NC-CAH diagnosis and obtained three births.

Ten of the 95 patients wanting pregnancy failed to become pregnant. In three couples, the husband had either azoospermia (n = 2) or severe oligoasthenoteratospermia (n = 1). Three other patients were 38–44 yr old when they consulted. Four women were lost to follow-up after a few months.

Miscarriages

The results of univariate analysis of the clinical and hormonal characteristics show that the rate of miscarriages was 26.3% for pregnancies occurring without glucocorticoid treatment and far higher than the rate of miscarriages (6.5%) for pregnancies obtained with glucocorticoid ($P < 0.01$). When we compare the rate of miscarriages in pregnancies occurring spontaneously without any type of treatment or with glucocorticoid treatment alone, it was 25.2 and 4.8%, respectively ($P < 0.001$) (Table 2). In addition, a past history of consultation for infertility was 3 times more frequent in the group with (30.8%) than in the group without (11.9%) miscarriages. However, the percentage of patients with menstrual disorders was 32.8% in those who presented miscarriage and

TABLE 3. Clinical and hormonal characteristics of patients (n = 85) who obtained pregnancy without or with glucocorticoid treatment (Tt)

Patient characteristics	Patients obtaining Pregnancy without glucocorticoid Tt (n = 49)	Patients obtaining Pregnancy with glucocorticoid Tt (n = 36)	*P*
Hirsutism	63.2%	83.3%	0.05
Menstrual cycle disorders	36.7%	58.3%	NS
Premature pubarche	2%	8%	NS
Infertility	14%	19.4%	NS
Age at diagnosis (yr)	32 ± 10	21 ± 6	<0.05
BMI (kg/m^2)	24.9 ± 5.6	23.4 ± 2.7	NS
Age at first pregnancy (yr)	26.8 ± 4.0	29.4 ± 4.3	NS
Basal 17OHP (ng/ml)	10.6 ± 12.5	11.2 ± 9.8	NS
Post-ACTH 17OHP (ng/ml)	45.4 ± 23.2	44.0 ± 26.0	NS
T (ng/ml)	0.75 ± 0.48	0.99 ± 0.46	<0.05
A (ng/ml)	3.8 ± 2.9	4.4 ± 2.4	<0.05
Basal FSH (mIU/ml)	4.7 ± 1.7	4.0 ± 1.5	NS
Basal LH (mIU/ml)	5.3 ± 4.2	4.7 ± 3.2	NS
Mutations			
Mild/mild	28%	29%	NS
Mild/severe	67%	67%	NS
Severe/severe	5%	3%	NS

BMI, Body mass index; NS, not significant.

38.4% in patients who did not have miscarriage event, which is not significantly different. In multivariate logistic regression, the risk of miscarriage in the absence of glucocorticoid treatment was significantly higher than with treatment (odds ratio 4.5, 95% confidence interval 1.43–14.2).

Cumulative incidence of pregnancies and TTP

As concerns the cumulative incidence of pregnancies for the 95 women wanting pregnancy, data were available for 121 pregnancies; 60.9% of the attempts were successful within 3 months, 66.9% at 6 months, 75.9% at 12 months, and 85.9% within 24 months. Among the 85 women who conceived, the cumulative pregnancy incidence was 73.5% at 6 months and 83.5% (101 pregnancies) at 12 months or less. These 101 pregnancies obtained during the first year led to 88 live births (82%).

Effect of glucocorticoid treatment on menstrual cycles and plasma androgen levels

Data on menstrual cycles before and with glucocorticoid treatment were available for 48 patients who received an average dose of 17.5 ± 7.5 mg hydrocortisone (0.73 ± 0.3 mg/kg/m^2); in the 38 patients of this group who presented with a- or oligomenorrhea before treatment, 27 obtained regular menstrual cycles with hydrocortisone treatment. Among the 11 patients presenting with amenorrhea before treatment, only three still had amenorrhea with treatment. Plasma T and A levels were available before and with glucocorticoid treatment in 55 patients. Plasma T decreased from 0.98 ± 0.49 to 0.45 ± 0.20 ng/ml and A from 4.65 ± 2.66 to 2.02 ± 1.68 ng/ml ($P < 0.001$) (Fig. 1).

Risk of a classical form of CAH in the children of NC-CAH patients

Among the 141 live births, two female infants presented a classical form of CAH: one salt wasting form (child 1) and one simple virilizing form (child 2). Child 1 was a compound heterozygote for two severe mutations: one large heterozygous 30-kb deletion inherited from her father and the complex rearrangement of the Reunion island (17) from her NC-CAH mother, herself a compound heterozygote for this mutation and V281L. At birth, she presented clitoromegaly, labia majora fusion, and vagina opening in the urethra at genitography. In addition, she had a salt-wasting syndrome. Child 2 was also a compound heterozygote for two severe mutations: the 290-13 A/C > G mutation in intron 2 inherited from her father and the I172N mutation from her NC-CAH mother, who also had the P30L mutation. At birth, child 2 presented posterior fusion of the labia majora.

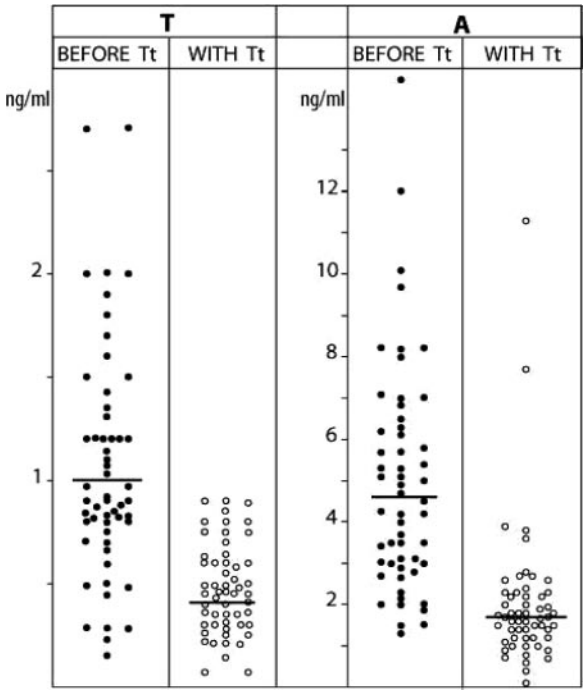

FIG. 1. Plasma T and A before and with hydrocortisone treatment (Tt).

These two pregnancies occurred before the diagnosis of NC-CAH was made in the mothers. It was the observation of external genitalia anomalies that led to the diagnosis of classical CAH in the children and the subsequent investigation of their mothers.

Discussion

Contrasting with numerous data on classical CAH, fertility in NC-CAH has been rarely studied (13–15). We therefore evaluated fertility in a group of 190 NC-CAH patients (161 index cases + 29 affected female relatives). Subfertility seems to be relative because 187 pregnancies, 110 without and 77 with glucocorticoid treatment, occurred in 85 of the 95 women (90.5%) who wanted pregnancy, leading to 141 births. These results are in agreement with previous reports (13–15). Birnbaum and Rose (13) observed 12 pregnancies among 22 NC-CAH patients desiring pregnancy. In the series reported by Feldman *et al.* (14), among 20 patients wanting pregnancy, 10 had conceived before the diagnosis and nine after NC-CAH diagnosis and after hydrocortisone treatment.

These results contrast with those concerning fertility in the classical form of CAH. The earliest studies reported a pregnancy rate of 0–10% in women with the salt-wasting form and 33–50% in the simple virilizing form (5–8). Some recent studies are more optimistic (12, 23, 24), whereas Gastaud *et al.* (9) observed a birth rate of 17% *vs.* 65% in a control population. Subfertility reported in the

classical form of CAH is related to several causes: morphological anomalies of genitalia due to precocious and severe virilization (5, 25); disturbed personal body image leading to psychological difficulties in social and sexual relationships (6, 12); and androgen oversecretion, which impairs hypothalamo-pituitary function during the peri- and postpubertal periods (14).

In NC-CAH, subfertility is mild compared with the classical form and seems to be mainly due to hormonal imbalance. Menstrual cycle or ovulation disorders observed in NC-CAH women who consulted for infertility were in most cases corrected by hydrocortisone treatment, which led to simultaneous lowering of plasma T and A levels and rapid occurrence of pregnancy. It has been suggested that the tonic oversecretion of androgens that may be aromatized to estrogens results in continuous steroid feedback and thus loss of gonadotropin cyclicity, leading to anovulation or dysovulation (26). Such persistent adrenal androgen oversecretion and subsequent conversion to estrogen can augment pituitary sensitivity to LHRH and increase LH release, as reported by Felman *et al.* (14) and as observed in our patients. This may lead to ovarian dysfunction, with a- or dysovulation mimicking the polycystic ovary syndrome (27–29). Adrenal androgens may also directly inhibit folliculogenesis by a negative effect on aromatase activity in granulosa cells (30). In addition, the possible persistence of higher-than-normal plasma progesterone levels may have an inhibitory effect through two mechanisms: by altering the rhythm and amplitude of GnRH pulses by an inappropriate feedback at the hypothalamo-pituitary level and modifying endometrial development during the follicular phase, thus altering nidation capacity.

The inhibitory effect of androgens on the pituitary-ovary axis is confirmed by the normalization of menstrual cycles subsequent to the suppression of adrenal androgen oversecretion by glucocorticoid treatment. This may also explain why the patients in our series who needed hydrocortisone treatment to obtain pregnancy were diagnosed with NC-CAH at a younger age and more frequently had hirsutism and higher plasma androgen levels than those who conceived without hydrocortisone.

In our cohort of 190 NC-CAH patients, 12% of the women consulted for infertility, which was secondary in 2.6%. This rate is comparable with that observed in the general French population (14–15%) (31). Mean age at first pregnancy in our patients was 27.9 ± 4.4 yr, similar to the age in the general French population. The voluntary abortion rate was 0.06 (0.52 in the general population) (32). It should be noted that voluntary abortion was mainly performed in pregnancies initiated in the absence of glucocorticoid treatment and was possibly related to careless contraception and the uncertainty of these women concerning their fertility in a context of frequent menstrual cycle disorders.

The cumulated pregnancy incidence rate for the whole group of 95 patients wanting pregnancy was 66.9% at 6 months and 75.9% at 1 yr. That is slightly less than in the general population (80 and 92%, respectively) (33–35). However when considering the 85 patients who conceived, the cumulative incidences of all pregnancies (83.5%) and pregnancies resulting in birth (82%) at 1 yr or less are similar to those in the general population (23).

The rate of miscarriage is 19.4% when abortions are excluded. We do not have a control group, but this rate is higher than the 10–15% rate reported in the general population (34, 35). The high percentage of miscarriages in our patients may reflect the poor quality of ovulation and/or the existence of an inadequate corpus luteum with its hormonal consequences on the uterine environment. The most striking observation is the higher number of miscarriages in pregnancies occurring without compared with glucocorticoid treatment (26.3 *vs.* 6.5%; Table 2). The difference remains marked when spontaneous and glucocorticoid only-treated pregnancies are compared (25.2 *vs.* 4.8%), which makes it possible to distinguish between glucocorticoid and ovulation inducer effects. A higher rate of miscarriages in the absence of glucocorticoid treatment has previously been suggested. Feldman *et al.* (14) reported six miscarriages in 18 pregnancies (33%) occurring in 10 NC-CAH patients before diagnosis, and none among pregnancies obtained on hydrocortisone treatment. Moran *et al.* (15) observed a markedly lower rate of miscarriages (25.4 *vs.* 6.2%) after NC-CAH diagnosis among 107 patients and 206 pregnancies. However, in the study by Moran *et al.*, some patients received glucocorticoid before NC-CAH diagnosis and some others ovulation inducers after NC-CAH diagnosis, which may dilute the specific glucocorticoid effect.

Hydrocortisone is considered to be an appropriate treatment (19, 36), and a twice-a-day hydrocortisone dose schedule seems to be a reasonable compromise between effectiveness and simplicity (37). The clinical criteria for therapeutic efficacy are regular cycles and a diphasic basal body temperature curve; the biological criteria consist of a fall in plasma T and A values to normal levels (14). However, it appears that a fixed glucocorticoid dose cannot accurately replace normal varying adrenal cortisol secretion, which is essentially designed to respond to stress. Patients receiving a standard dose of hydrocortisone are subjected to alternate periods of under- and overtreatment, depending on their stress level. Androgen peaks can occur during the periods of undertreatment and probably can impair normal cyclic gonadotropin axis function (38).

When hydrocortisone therapy does not lead to normal gonadotropin function, clomiphene citrate may be proposed. Clomiphene's antiestrogen action interrupts the positive feedback permanently exerted on the hypothalamopituitary axis by estrogens produced from aromatization of adrenal androgens, thereby permitting FSH output, follicular maturation, and a rise in plasma estradiol, which is thus able to trigger a LH peak and ovulation (39). Once pregnancy occurs, hydrocortisone is usually maintained at a dose of 20–25 mg/d. Finally, the rate of preterm deliveries was 4.3%, which is close to the rate of 5–6% observed in the french population (40) and did not differ with or without glucocorticoid treatment.

In conclusion, subfertility in NC-CAH female patients seems to be relative. It is not a frequent reason for consultation; in addition, when present, it is easily resolved. In our NC-CAH cohort, 85 of the 95 patients who desired to become pregnant obtained 187 pregnancies, 110 before and 77 with glucocorticoid treatment. Hydrocortisone seems to be the appropriate treatment; it lowers adrenal androgen secretion, normalizes menstrual cycles, and reduces the incidence of miscarriages. Considering the classical form of CAH diagnosed at birth in two children of our NC-CAH patients and the high frequency of *CYP21A2* mutations in the general population (15), it is essential to genotype the partner of NC-CAH patients with one severe mutation to offer genetic counseling.

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