

# Genetic and Clinical Spectrum of Aromatase Deficiency in Infancy, Childhood and Adolescence

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## Key Words

Androgen excess · Aromatase deficiency · Estrogen replacement · Hypothalamo-pituitary-gonadal axis · Insulin insensitivity

## Abstract

**Introduction:** cP450aromatase deficiency provides clues for the understanding of the role of aromatase in prepubertal and pubertal human health and disease. Placental aromatization of androgens protects the female fetus against the virilizing action of fetal androgens. After birth, the dual effect of aromatase deficiency, excessive androgens, and insufficient estrogens is responsible for a variable clinical picture. Nineteen cases of aromatase gene (*CYP19*) deficiency have been reported. **Phenotype:** Phenotype is dependent on sex and age. In newborns, aromatase deficiency should be considered in the etiology of 46,XX DSD, after ruling out congenital adrenal hyperplasia. In prepubertal aromatase deficient girls, high levels of ovarian androgens and gonadotropins facilitate the formation of ovarian cysts. Bone mineralization can be affected and bone aging is delayed. In pubertal girls, there is poor sexual development and abnormal virilization. The phenotype may be variable according to enzyme activity level. Insulin sensitivity may be abnormal in

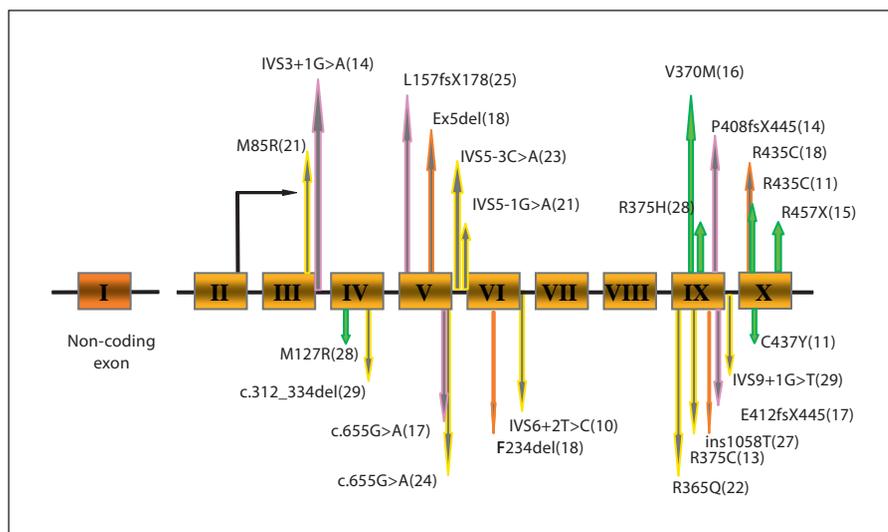
both men and women. Finally, aromatase might also play a role in the regulation of testicular cell mass in the newborn testis. **Conclusion:** Adequate interpretation of clinical data should lead to the analysis of the *CYP19* gene for diagnostic confirmation and implementation of appropriate management.

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Several molecular *CYP19* gene alterations associated with cytochrome P450aromatase (cP450arom) deficiency have been described in both females and males. They have contributed considerably to the analysis of cP450arom activity in different tissues, influencing sexual differentiation, patterns of gonadotropin secretion by the hypothalamic-pituitary axis, reproduction capacity, lipid metabolism, insulin sensitivity, and skeletal maturation and growth. Several reviews of this rare disorder have been published [1–4] addressing mainly the clinical picture in affected adult men [4–6].

This review discusses clinical, biochemical and molecular aspects of human cP450arom deficiency during childhood in both sexes and in affected pubertal subjects.

**Fig. 1.** The human *CYP19* (cP450arom) gene, located in the 21.2 region on the long arm of chromosome 15 (15q21.2). Its regulatory region contains at least 10 distinct promoters regulated in a tissue- or signaling pathway-specific manner. The coding exons, namely exons II–X, are shown, along with the location of the reported mutations.



### cp450arom Characteristics and Reported Human Mutations

cP450arom is the enzyme that catalyses the synthesis of estrogens from androgens. Therefore, the activity of this enzyme complex affects both androgen metabolism and estrogen synthesis. The biological importance of the aromatase complex is related not only to its role in the synthesis of estrogens, but also to its potential influence on the balance of the androgen-estrogen ratio in different tissues. The aromatase complex is expressed in the endoplasmic reticulum of cells, and consists of 2 components: cP450arom and a ubiquitous flavoprotein, NADPH-cytochrome P450 reductase. Even though androstenedione and testosterone are the most common and the most physiologically important substrates, 16OH-dehydroepiandrosterone sulfate (16OH-DHEAS) arising from fetal liver hydroxylation of fetal adrenal DHEAS is an important substrate for placental estriol synthesis during pregnancy, both in humans and higher primates [1].

In humans, cP450arom appears to be the product of a single gene – *CYP19* – located on chromosome 15q21.1. The protein-coding sequence is contained within 9 exons (2–10), that span approximately 35 kb of DNA [7]. The translation, initiation and termination codons are present between exons 2 and 10. However, there are multiple first exons that are involved in tissue-specific expression. These exons, that are not translated, generate alternative splicing in such a fashion that the coding region, and hence the protein sequence, is conserved in every tissue. Several promoters are found within a 90-kb region up-

stream of the coding region [7]. The most highly conserved region (the core region), defined by a 3-dimensional model of cP450arom, consists essentially of a 4-helix bundle, 2  $\beta$  sheets and the heme-binding region.

cP450arom is expressed in a number of tissues, including the syncytiotrophoblast layer of the placenta, gonads, adipose tissue, bone, brain (including the hypothalamus, hippocampus, and amygdala), coronary arteries, and various fetal tissues, such as liver, skin, intestine, testis and ovary [8].

*CYP19* gene expression is subjected to multifactor regulation by a diverse group of hormones and factors that differ markedly between tissues. Thus, a strict control over tissue-specific expression is necessary for proper regulation of estrogen synthesis during fetal development, as well as during post-natal life.

Since aromatase deficiency was first described by Shozu et al. in 1991 [9], 19 cases have been reported (11 females [9–21] and 8 males [13, 22–29]). Molecular defects, in vitro aromatase activity of mutants, and clinical phenotype in subjects who were of pre-pubertal or pubertal age at diagnosis are described in table 1. Variations in phenotype are compared with in vitro functional rearrangements of mutants. Table 2 shows molecular defects and in vitro aromatase activity in subjects who were adults at diagnosis. Curiously, all reported adult subjects were males in whom the main phenotypic characteristics are tall stature and varying degrees of mild testicular hypofunction. Figure 1 shows a cartoon of the cP450arom gene with its exons, depicting the main mutations described so far in humans.

**Table 1.** Mutations of the CYP19 gene reported in subjects who were of pre-pubertal or pubertal age at diagnosis

Sex [ref.]	Gene mutations	Molecular defect	In vitro aromatase activity	Phenotype
Female [10]	Homozygous (hm) IVS6 + 2T>C	Mutation at consensus 5' splice site (5'ss) in intron 6 resulting in the use of a cryptic site downstream and in the in-frame incorporation of 87 nucleotides. A protein with additional highly hydrophobic 29 amino acids will be generated.	Less than 0.3% of WT control. The extra aa might induce conformational changes and reduction of activity.	Ambiguous genitalia at birth.
Female [11]	Compound heterozygous: R435C C437Y	R435C: Cysteine instead of arginine at codon 435, exon 10. Highly conserved residue within the heme-binding region among cP450s. C437Y: tyrosine instead of cysteine at position 437, exon 10. Cysteine 437 is the critical cysteine in the heme-binding domain and is the best conserved residue in all cP450 proteins [8].	R435C mutant protein showed approximately 1.1% of WT activity. C437Y mutant protein was inactive.	Ambiguous genitalia at birth. Puberty absent. Delayed bone age, ovarian cyst and virilizing signs at pubertal age.
Female [13]	hm R375C	Exon 9 mutation, a cysteine instead of an arginine	0.2% of WT activity. Protein modeling studies suggest that the affected region may affect the substrate access channel to the membrane.	Ambiguous genitalia at birth. Puberty absent. Ovarian cyst and virilizing signs at pubertal age.
Female [15]	hm R457X	Point mutation in exon 10 generating a premature stop codon.	Not determined.	No detailed phenotype reported.
Female [14]	Compound heterozygous: P408fsX445 IVS3+1G>A	P408fsX445: A base pair deletion (C) in codon 408 (exon 9). A frameshift occurs resulting in a nonsense codon. IVS3+1G>A: G to A change at the canonical donor splice site in intron 3. The splice site is ignored, and a stop codon arises 3 bp downstream. No active transcript was found.	Not determined. However, no aromatase activity is expected.	Ambiguous genitalia at birth. Ovarian cysts during infancy and childhood. Low BMD at 3.5 years of age.
Female [16]	hm V370M	G to A transition in exon 9 that results in the amino acid change valine to methionine.	Not determined.	Ambiguous genitalia at birth.
Female infant [17, 19]	Compound heterozygous: C655G>A E412fsX445	E412fsX445: a bp deletion (A) in glu 412 (exon 9) causing a frame shift generating a stop codon 98 bp downstream. Subsequently, a truncated protein with an altered heme-binding domain might be generated. C655G>A: G to A at the consensus donor splice site in exon 5 to intron 5 junction causing in-frame exon 5 skipping and a putative inactive protein lacking amino acids 151–209 encoded by exon 5 [11].	The peptide expressed from E412fsX445 is expected to be completely inactive as the frame shift altered the highly conserved heme-binding domain. The protein without amino acids 151–209 encoded by exon 5 does not have aromatase activity. As the –E5 mRNA might be expressed as a splice variant in normal tissues, some +E5 transcripts from the C655G>A allele might explain the partial aromatase deficiency phenotype.	Ambiguous genitalia at birth. Ovarian cysts during infancy, childhood and puberty. Delayed bone age during childhood. Spontaneous breast development at 7.7 years old. Virilizing signs at pubertal age. Normal BMD during infancy, childhood and puberty. Insulin resistance since 7.2 years of age and Type 2 diabetes during follow-up.
Pubertal female [18]	hm R435C	Cysteine instead of arginine at position 435 in exon X.	0.7–1.5% of WT activity. The arginine at position 435 forms part of the absolutely conserved heme-binding motif and is absolutely conserved.	Ambiguous genitalia at birth. Ovarian cysts and delayed bone age. Spontaneous breast development.
Female infant followed through puberty [18]	hm F234del	Deletion of 1 of 2 phenylalanines at position 234–235 in exon VI. This pair of amino acids is highly conserved in cP450 aromatase of other species.	16–19% of WT activity. Substrate binding seems to be largely unaffected but maximal enzymatic activity is reduced. The resultant protein was found to be completely inactive.	Ambiguous genitalia at birth. Low androgens levels in a female infant. Spontaneous puberty, delayed bone age and ovarian cysts in a pubertal female. Severe virilization (raised as male).

**Table 1** (continued)

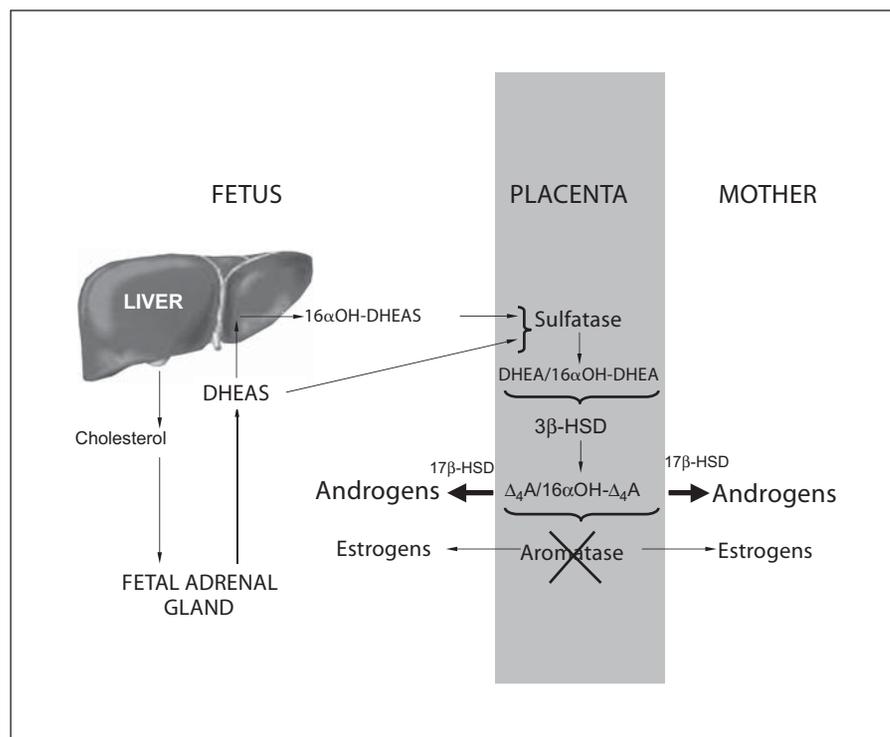
Sex [ref.]	Gene mutations	Molecular defect	In vitro aromatase activity	Phenotype
Pubertal female [18]	hm EX5del	600 bp deletion at genomic DNA. This deletion is predicted to remove exon 5 resulting in the in-frame deletion of 59 amino acids.	Not determined. However, no aromatase activity is expected.	Labial fusion at birth. Delayed puberty and bone age. Small ovaries. Low androgen levels. Moderate dyslipidemia.
Female [21]	Compound heterozygous: M85R IVS5-1G>A	M85R: arginine instead of methionine at codon 85 (exon 3).	The protein expressed from the M85R allele showed no activity.	Ambiguous genitalia at birth. Puberty absent and bone aging delayed. Virilizing signs at pubertal age.
Male infant [25]	hm L157fsX178	C base deletion in exon 5 causing a frame shift and a premature stop codon after 21 codons.	Not determined. However, the resultant peptide does not contain the substrate binding pocket (I helix), the electron accepting site and the heme-binding site [21].	Normal genitalia at birth.

BMD = Bone mineral density; WT = wild type.

**Table 2.** Mutations of the CYP19 gene reported in subjects who were adults at diagnosis

Sex [ref.]	Gene mutations	Molecular defect	In vitro aromatase activity
Male [13]	hm R375C	Exon 9 mutation resulting in cysteine instead of arginine at codon 375.	Activity of the mutant protein was 0.2% of wild type. Protein modeling studies suggest that the affected region may be important in anchoring the region of the protein proximal to the substrate access channel to the membrane.
Male [22]	hm R365Q	G to A mutation at base pair 1094 in exon 9 resulting in glutamine instead of arginine at codon 365.	Activity of the mutant protein was 0.4% of wild type.
Male [23]	hm IVS5-3C>A	C to A transition at bp -3 at the splicing acceptor site in exon VI. Exon VI is completely excised leading to a frame shift and premature stop codon 8 nucleotides downstream from the end of exon 5.	The resulting peptide most likely will not be processed, but even if it were, it would not result in a functional aromatase because it lacks the substrate-binding pocket, the electron-accepting site and the heme-binding site [23].
Male [27]	hm Ins1058T	T insertion at nucleotide 1058 in exon 9. This insertion leads to a frame shift and the appearance of a premature stop codon 24 codons later.	The protein is expected to be inactive as the heme and steroid-binding domain would not be present.
Male [24]	hm c655G>A	G to A transition in the last nucleotide in exon 5. The mutated DNA will generate an mRNA that includes the intron 5 sequence which contains an in-frame stop codon 30 bp downstream of the splice junction.	Not determined. A truncated and inactive protein lacking the heme-binding domain would be expected.
Male [28]	Compound heterozygous: M127R R375H	M127R: Arginine instead of methionine at codon 127 (exon IV). R375H: histidine instead of arginine at codon 375 (exon IX).	Not determined.
Male [29]	Compound heterozygous: c.312_334del IVS9+1G>T	c.312_334del: 23-bp deletion in exon 4 that would be expected to cause a frame shift with a premature stop codon at nucleotide 361 in exon 4. IVS9+1G>T: point mutation in the first nucleotide of intron IX that would lead to aberrant splicing of the mRNA.	Not determined. A truncated and inactive protein lacking the heme binding domain would be expected from the c.312_334del allele.

**Fig. 2.** Compartmentalization of sex hormone metabolism in the fetoplacental unit. DHEAS and  $16\alpha\text{OH-DHEAS}$ , synthesized in large amounts by interactions of fetal adrenal gland and liver, are transferred to the placenta. Here, after sulfate hydrolysis and dehydrogenation, active androgens are produced. One important role of aromatase is to neutralize these androgens by converting them into estrogens. In aromatase deficiency, androgens accumulate and virilize the female fetus and the mother.



### Impact of Aromatase Deficiency

The clinical characteristics reported in female patients with aromatase deficiency during pre-puberty and puberty, as well as estrogen replacement therapy and the impact of insulin sensitivity, are reviewed in this section. For comparison, the only male infant reported is also discussed.

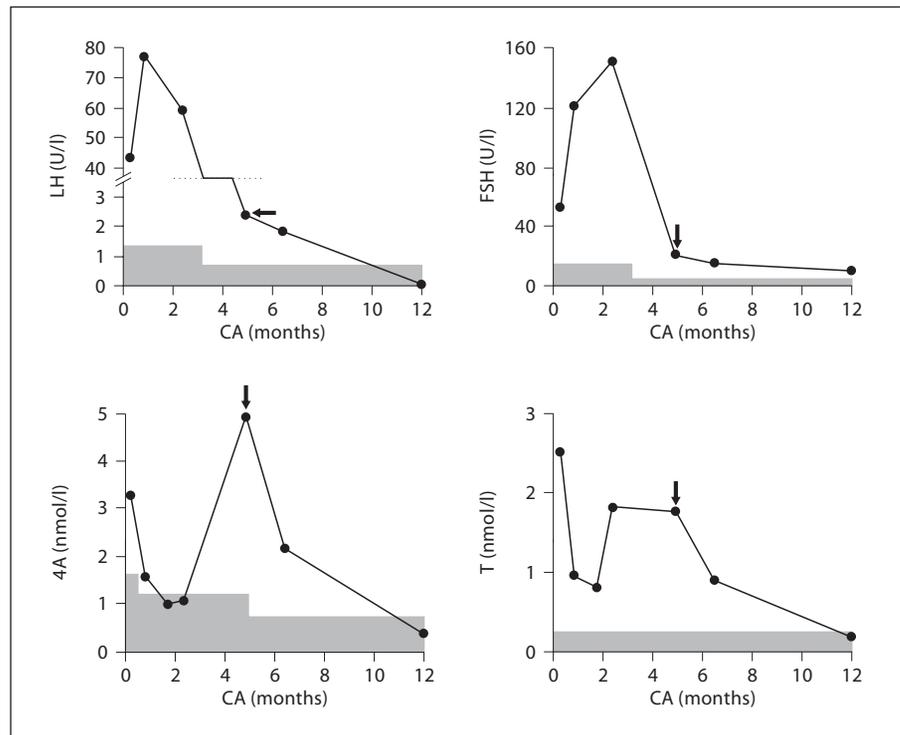
#### Female Newborns

The active human placental aromatization of androgens protects the fetus against the virilizing action of fetal androgens (fig. 2). In the female fetus, this is particularly important to avoid the effect of androgens on differentiation of external genitalia. It also plays a role in the central nervous system, in programming the brain during fetal and neonatal life for noncyclic hypothalamic GnRH function, as well as male sexual identity and behavior. In most female cases of aromatase deficiency described, ambiguous genitalia with various degrees of masculinization of the external genitalia have been reported. As expected, gonads were nonpalpable and female internal genitalia differentiation was not affected [9, 11, 13, 17–19, 21]. In addition, signs of maternal virilization (acne, deep voice, clitoris enlargement) during pregnancy might occur

and this should alert obstetricians to the possibility of this diagnosis. After delivery, these signs and symptoms disappear gradually [9, 13–14, 17–19, 21]. Onset of labor has been described as spontaneous [9, 14] and babies were born full term with adequate weight for gestational age [9, 11, 13–14, 17].

In a newborn with 46,XX ambiguous genitalia, the diagnosis of congenital adrenal hyperplasia should be ruled out first, because of its high frequency. However, detecting signs of virilization in the mother is useful clinical information in aromatase deficiency, as it does not occur in congenital adrenal hyperplasia.

An endocrinological profile has been described in some affected females during the first month of postnatal life. High androgen levels in cord serum have been reported, but serum levels of androgens returned rapidly to normal after delivery in some cases [9, 12, 14]. The serum patterns of basal gonadotropins during this period have been reported in only 1 aromatase-deficient girl by Belgorosky et al. [17]. In this patient, serum LH and FSH levels were extremely high, resulting in high serum androgen levels. This abnormal serum gonadotropin pattern might reflect a central change in the activity of the GnRH pulse generator and/or an effect at the pituitary level, presumably induced by the increment of androgens



**Fig. 3.** Serum LH, FSH, androstendione (4A) and testosterone (T) during the first year of life in an aromatase-deficient girl. The figure shows extremely high serum LH, FSH, 4A and T during the first 5 months of life, followed by a marked decrease thereafter. However, serum FSH never reached normal levels. A large ovarian cyst was removed at 5 months of age (arrow) coinciding with a huge increase of 4A and T. Hormone values decreased after cyst removal. Shaded areas depict normal ranges for the 4 hormonal determinations. Copyright © 2003 The Endocrine Society.

and/or local estrogen deficiency during fetal and neonatal life (fig. 3).

#### Female Infancy and Childhood

A clinical phenotype, including changes in the hypothalamic-pituitary-gonadal axis, ovarian cyst development, impact on growth, skeletal maturation, and bone homeostasis, as well as changes in insulin sensitivity and lipid profile, has been reported in affected girls.

#### Hypothalamic-Pituitary-Gonadal Axis

The hypothalamic-pituitary-gonadal axis has been studied in 5 affected girls with aromatase deficiency during infancy and childhood [11, 14–15, 17, 18].

In one girl, reported by Belgorosky et al. [17], serum basal FSH and LH levels dropped dramatically between 2 and 6 months of age, but whereas serum basal FSH levels remained clearly high, serum basal LH levels decreased gradually to reach normal levels at the end of the first year of life. On the other hand, during the same age period, basal levels of serum androgens remained high and multiple follicular cysts developed in both ovaries (fig. 3). However, serum androgen levels dropped after a large ovarian cyst was removed. In another girl, reported by Mullis et al. [14], basal serum FSH levels and the FSH

response to GnRH were elevated at 2 months of age, but normal basal and GnRH-induced LH levels were found.

During childhood, both serum basal and GnRH-induced FSH levels remained clearly elevated in these patients [14, 17]. Furthermore, patients reported by Conte et al. [11] at 17 months of age, and by Lin et al. [18] at 2.3 years of age, showed high basal and LHRH-induced serum FSH levels. Therefore, the persistence of high serum basal FSH levels observed in these affected girls suggests that the minimal amounts of normal prepubertal circulating estrogens are enough to restraint pituitary FSH. Serum basal LH levels were within the normal range in all reported patients, but a hyper-response after GnRH stimulation was observed. It could be speculated that a prolonged effect of androgens or lack of estrogens during childhood might result in an irreversible maturation of the GnRH pulse generator [30].

#### Ovarian Cyst Formation

Large hemorrhagic cystic follicles have been described in aromatase-deficient girls during infancy and childhood [11, 14, 17] and in ArKO female mice [31]. It is remarkable that the histopathologic analysis was consistent with that of the polycystic ovary syndrome. It has been proposed that chronic exposure to abnormally high levels

of LH stimulates ovarian cyst development [32, 33]. However, female mice that are homozygous for a targeted disruption of the FSH  $\beta$ -subunit gene exhibit an approximately 5-fold increase in serum LH, but do not show signs of enlarged cystic follicles in the ovary [34], suggesting there is a role for FSH in this process as well. In the rat, granulosa cells of primary, secondary and mature follicles show intense androgen receptor expression [35]. Weil et al. [36] showed that testosterone increased follicular FSH receptors and it has been suggested that androgens, such as those present in polycystic ovary syndrome, promote follicular growth by amplifying the FSH effect. It has been proposed that this partially explains the enhanced responsiveness to gonadotropin stimulation noted in women with polycystic ovary syndrome. Therefore, we have speculated that in aromatase-deficient prepubertal girls, an amplification of FSH signaling might occur in the presence of high intraovarian androgen production and that this mechanism could be involved in the development of ovarian follicular cysts.

The usefulness of estrogen treatment during infancy and childhood in affected female patients is not clear. Mullis et al. [14] reported that low doses of estradiol in a 3-year-old affected girl resulted in normalization of serum gonadotropins, regression of enlarged ovaries and improvement in bone mineral density (BMD). However, plasma FSH returned to pretreatment levels and ovarian size increased shortly after of estrogen withdrawal. Although the ovary is not quiescent during the prepubertal period, and a minipuberty has been described during the first 3 months of age in normal girls, there is little experience in potential side effects of chronic low-dose administration of estradiol during early prepuberty. Therefore, to prevent the risk of torsion of enlarged ovaries and ovarian cysts during infancy and early childhood, long-term treatment with a GnRH analog may be considered as a therapeutic option. However, an undesired effect of lack of gonadal hormones on BMD during infancy and early childhood has not been clearly established.

#### Impact on Growth, Skeletal Maturation and Bone Homeostasis

Delay in bone maturation has been reported in aromatase deficiency [17]. Particularly, a delay in skeletal maturation was clearly evident in late prepuberty. Furthermore, an advance of bone age under estrogen replacement therapy has been reported [13], confirming the critical role of estrogens on skeletal maturation during prepubertal years.

It is accepted that estrogens are important in preserving adequate BMD [37]. However, data on the role of estrogens on bone mineralization during childhood are scarce. In the patient reported by Mullis et al. [14], a low BMD was found. Moreover, BMD improved after treatment with low doses of estrogens, suggesting that minimal quantities of estrogens are also required to preserve mineral bone acquisition in early childhood. However, in the case reported by Belgorosky et al. [17], BMD was normal during the entire follow-up. The explanation could be that in this particular case a partial cP450arom deficiency, secondary to a mutation at the consensus donor splice site in the exon 5/intron 5 junction was found (table 1). Therefore, some expression of cP450arom protein might be expected, which may be enough to maintain a normal mineral bone acquisition, at least during childhood.

#### Female Puberty

##### Classic Phenotype

Classically, in complete aromatase deficiency [11, 13, 18, 21] affected females show absence of the onset of puberty, with lack of breast development and primary amenorrhea, as well as excessive virilization. The pubertal spurt is absent and bone aging delayed. Basal and GnRH-induced LH and FSH levels are elevated. Serum androgens (testosterone and androstenedione) levels are high compared to normal values for age and stage of sexual development, whereas serum estradiol levels are extremely low. This hormonal pattern supports the concept of a primary role of estrogen, not replaced by androgens, in the feedback mechanism of gonadotropin secretion within the hypothalamo-pituitary-gonadal axis in females. Multiple bilateral ovarian cysts are also seen on pelvic ultrasound. In the case reported by Morishima et al. [13], several cystic masses with multiple lobules were found on abdominal exploration.

##### Variable or Non-Classic Phenotypes

Recently, Lin et al. [18] have reported the presence of variable, or non-classic, phenotypes in 2 affected females. A 13.5-year-old patient showed findings compatible with the complete form of aromatase deficiency, except for the presence of spontaneous breast development that did not progress further than Tanner stage 2, and a pubertal size uterus with an endometrial stripe. The second female studied had spontaneous breast development that progressed to Tanner stage 4 at 14 year of age. Since she was raised as a male and reported male gender identity, role and orientation, salpingo-oophorectomy and hysterectomy

tomy were performed in an attempt to reduce sex hormone secretion and arrest breast development (table 1).

The patient described by Belgorosky et al. [17], unexpectedly started with spontaneous breast development (Tanner stage 2) at the age of 7.7 years, and a slight elevation of serum estradiol levels was detected.

The cases described by Lin et al. [18] and Pepe et al. [19] illustrate that phenotypic variability may occur in aromatase insufficiency in humans. Low levels of residual aromatase activity may be associated with breast development and estrogen biosynthesis, specially when circulating androgen substrate concentrations are elevated allowing for a phenotype of partial cP450arom deficiency.

Although detailed psychosexual studies have not been reported, female psychosexual orientation has been observed in most 46,XX subjects, except for the patient published by Lin et al. [18]. It is unclear whether pre- and postnatal androgen exposure coupled with relative estrogen insufficiency, a unique feature of aromatase deficiency, is important in programming sexual orientation, or social and cultural influences predominate.

During puberty and adulthood, signs and symptoms of estrogen deficiency become evident. In pubertal females with aromatase deficiency, daily estrogen replacement therapy associated with a progesterone-like hormone not only resulted in breast development, menarche and maintenance of menses, growth spurt and fused epiphyses, but also in decreases in plasma concentration of gonadotropins and androgens with subsequent regression of ovarian cysts [11, 13, 18]. Thus, treatment confirmed the critical role of estrogens on the hypothalamic-pituitary-ovarian axis and on bone skeletal maturation. Although no data exist on bone mineral mass in affected females on estrogen replacement therapy, it is expected that estrogen treatment might be important to improve bone mineral accretion.

#### *Insulin Sensitivity and Lipid Profile*

Insulin resistance, abnormal lipid profile, as well as clinical features similar to the metabolic syndrome have been described in aromatase-deficient adult men. Improvement of these parameters with estrogen therapy has been reported [22–24, 29, 38]. However, in affected female patients, the relationship between carbohydrate metabolism, lipid profile and estrogen deficiency has not been clearly defined. There is only 1 available report, by Lin et al. [18], of an affected 14.6-year-old girl, who had a mild dyslipidemia and whose metabolic profile normalized under estrogen therapy.

Insulin resistance and glucose intolerance at the age of 9 years, associated with high serum androgen and FSH levels have been reported in an affected girl with early signs of puberty by Guercio et al. [39]. No response to estrogen replacement therapy or metformin treatment was observed. She developed Type 2 diabetes mellitus during follow up. Lipid profile, liver enzyme levels and liver ultrasound were normal. In this patient, GnRHa administration for 1 year resulted in marked decreases in gonadotropin and serum androgen level, but severe insulin resistance persisted. This lack of response to estrogen replacement and androgen withdrawal suggested that the metabolic abnormality was irreversible. The authors proposed that in females the androgen increase and/or lack of estrogens during fetal life might have altered the mechanism of fetal programming of insulin sensitivity.

#### *Aromatase Deficiency in an Affected Infant Boy*

Deladoëy et al. [25] reported the only affected infant boy described in the literature. His mother suffered from progressive virilization during pregnancy but at birth, as expected, his external genitalia were normal. In contrast to affected infant girls, hormonal studies carried out when he was 2 month old revealed that basal and GnRH-stimulated FSH levels were normal despite low serum estrogen concentrations, suggesting that estrogens are not involved in the regulation of FSH secretion in infant boys. Hence, a gender-specific negative feedback mechanism, at least during infancy and early childhood, has been suggested [25]. Moreover, in this aromatase-deficient boy, a normal serum inhibin B level for sex and age was found. Hence, it had been proposed that inhibin B might be the major contributor in the regulation of serum FSH secretion in normal infant males. The fact that in the affected boy free testosterone and androstenedione levels were very high at 2 weeks of postnatal life, followed by a decrease to the normal range during the first month of age suggests a role of cP450arom in fetal and newborn testicular function.

It has been described that cP450arom is highly expressed in fetal and neonatal human testes compared to testes of older prepubertal ages. Berensztein et al. [40] suggested that aromatase activity might play a role in modulating testosterone secretion; the high aromatase expression of human neonatal Leydig cells decreased at 2–4 months of age whereas peak testosterone secretion occurred during the postnatal testicular activation period.

Berensztein et al. [41] also found that during the newborn period, there is a vigorous increment in testicular

cell populations. This cell mass growth seems to be mainly mediated by decreased apoptosis. It has been postulated that these changes, taking place during the neonatal period, may be important to define testicular function in adults. The factors that modulate this growth are not known, but aromatase might play a role.

On the other hand, in aromatase-deficient adult males, testis volume has been reported to be variable. In a few patients, decreased motility and reduced numbers of spermatozoa have been observed on sperm analysis. Therefore, testis function in aromatase-deficient adults is not well defined.

### Final Comments

Patients with cP450arom deficiency represent another 'experiment of nature' that has proven to be a fertile source of knowledge of estrogen physiology and pathology, particularly on the roles of estrogens and bone mat-

uration, gonadotropin modulation in both sexes, ovarian cyst formation, testicular function and glucose and lipid metabolism. However, many questions remain to be answered, such as the possible existence of a window period for estrogen action in early life, in both sexes. In females, the increment of androgens and/or lack of estrogens during fetal life might be involved in a mechanism of fetal programming on insulin sensitivity and on the hypothalamic-pituitary-gonadal axis. In males, lack of local estrogen production in fetal or neonatal testes could alter adult testicular development and function.

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