Estrogen Deficiency in Men

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Abstract

Congenital estrogen deficiency in men is a rare disorder that remains overlooked and undermanaged till adulthood. Similarly, other genetic diseases causing congenital hypogonadism are rare and indirectly lead to estrogen deficiency during infancy and puberty if not recognized and treated. Apart from congenital, genetic forms, estrogen deficiency may occur as a consequence of hypogonadism and reduced androgen production. Several lines of evidence support the idea that estrogen deficiency may be detrimental for several male physiological functions, especially in aging. Among them, bone loss, osteoporosis, increase of fat depots, and sexual function may depend to a various degree from estrogen deficiency. At present, however, nosological data on estrogen deficiency in men are lacking. This chapter describes the pathogenesis and clinical manifestations related to estrogen deficiency and provides clinical advice on how to diagnose and treat both congenital and acquired forms of estrogen deficiency.

Keywords

Estrogen deficiency • Aromatase deficiency • Estrogen resistance • Male hypogonadism • Male osteoporosis • Estrogens in men

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Abbreviations	
ADT	Androgen deprivation therapy
BMD	Bone mineral density
DEXA	Dual energy X-ray absorptiometry
ED	Estrogen deficiency
ER	Estrogen receptor
FSH	Follicle-stimulating hormone
GnRH	Gonadotropin releasing hormone
IGF-1	Insulin-like growth factor 1
IIH	Isolated hypogonadotropic hypogonadism
LC-MS/MS	Liquid chromatography-tandem mass spectrometry
LH	Luteinizing hormone
PORD	P450 oxidoreductase deficiency
T	Testosterone

Contents

Introduction

Estrogen deficiency in men is still a poorly understood clinical entity. Genetic forms of estrogen deficiency are rare (Rochira and Carani 2009) and data on relative estrogen deficiency due to testosterone deficiency and the consequent fall of serum estrogens below the normal range are scanty (Trabado et al. 2011; Finkelstein et al. 2013; Giton et al. 2015). Furthermore, no nosological data on estrogen deficiency are available. In clinical practice, relative estrogen deficiency due to male hypogonadism is common, especially if compared to the genetic forms. Probably, not all hypogonadal men develop estrogen deficiency, the presence and severity of which depend from the degree of testosterone deficiency as well as from the individual production rate of estrogens. However, the real prevalence of relative estrogen deficiency is still not known. Since several years ago, estrogens in men were considered relatively unimportant and their physiological role has been

progressively disclosed only in recent years (Simpson and Santen 2015). For a comprehensive review on the milestones concerning the knowledge of estrogen role in men see (Rochira et al. 2005).

At present there is strong evidence that estrogens exert their pleiotropic actions on several tissues in men (Rochira et al. 2012) (see also ▶ Chap. 11, "Anabolic and Metabolic Effects of Testosterone and Other Androgens: Direct Effects and Role of Testosterone Metabolic Products" for details). In particular, the role of estrogens is crucial for the skeleton (Rochira et al. 2015; Almeida et al. 2017) and for male reproduction (Rochira et al. 2005, 2016) as well as for other important physiological functions (Rochira et al. 2012).

For these reasons, estrogen deficiency in men is receiving increasing interest in the last years (Finkelstein et al. 2013, 2016; Taylor et al. 2016).

Physiology of Estrogens in Men

Estrogen Synthesis and Action

Estrogens in men derive from the conversion of androgen thanks to the action of the enzymatic complex known as aromatase, which is responsible for the aromatization of the androgen A-ring (Simpson et al. 1999; Miller and Auchus 2011; Rochira et al. 2016) (see also ▶ Chap. 10, "Testicular Steroidogenesis" for further details) (Fig. 1).

As other steroidogenetic enzymes, aromatase belongs to the large family of cytochrome P450 oxidases, each sharing a conserved heme-binding domain and differing for a variable substrate-binding domain (Harada 1988). Aromatase is encoded by the *CYP19A1* gene, which consists of ten exons (Harada 1988; Harada et al. 1990). The translated coding region belongs to exons 2–9 that are not tissue specific, whereas the noncoding exon 1 differs among various tissues. The presence of different exons 1 explains how the expression of aromatase is regulated in a tissue-specific fashion (Harada et al. 1990, 1993; Simpson et al. 1997). Accordingly, the *CYP19A1* gene includes multiple tissue-specific promoters, which allows the aromatase transcript to be tissue-specifically spliced from the multiple alternative exons available for exon 1 (Harada et al. 1990, 1993; Simpson et al. 1997).

The primary transcript is spliced into various transcripts in a tissue-specific fashion as a result of tissue-specific promoters (Harada 1988; Simpson et al. 1997). Finally, the mature protein includes the sequences encoded by exons 2–10 (Simpson et al. 1994, 1997).

The main sources of estrogen in men are the testes, the liver, and the adipose tissue (Gruber et al. 2002; Barakat et al. 2016), including in particular the tissues that express the aromatase enzyme and are able to locally produce estrogens (e.g. testis, adipose tissue, brain, including the hypothalamus, bone, muscle, breast, liver) (Simpson et al. 1999; Gruber et al. 2002; Barakat et al. 2016).

Estrogen action occurs through binding to specific receptors belonging to the nuclear receptors superfamily (Gruber et al. 2002; Heldring et al. 2007). Estrogen

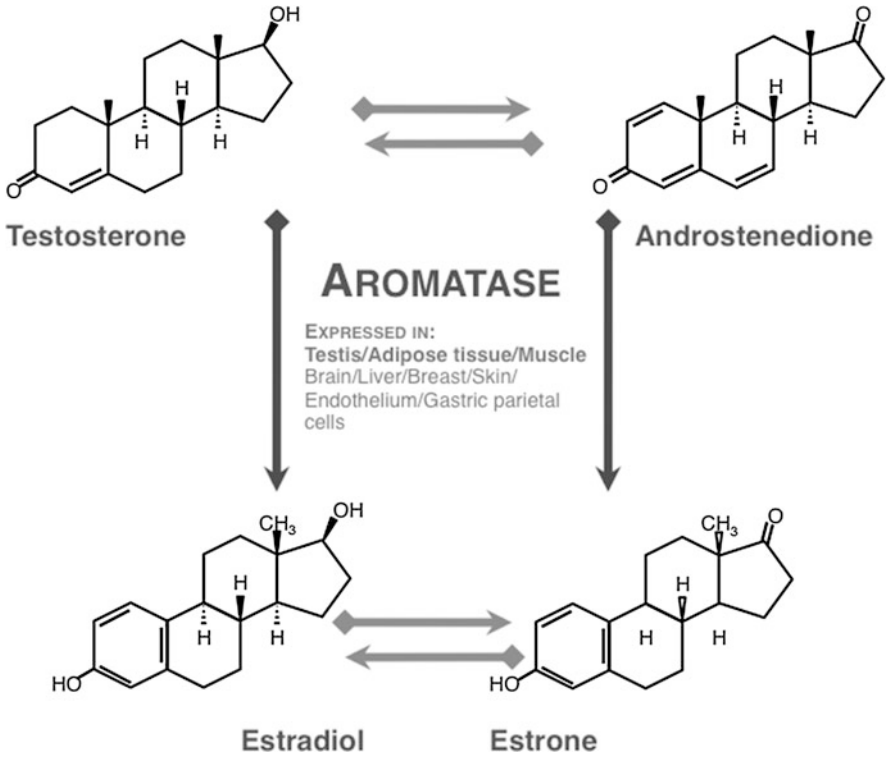

Fig. 1 Estrogen biosynthesis in men and main tissues expressing the aromatase enzyme

receptors are ligand-inducible transcription factors, which regulate the expression of target genes after hormone binding (Heldring et al. 2007). Two different types of intranuclear estrogen receptors (ERs) have been characterized insofar: the ER-alpha (ERα) and the ER-beta (ERβ) (Heldring et al. 2007). Furthermore, even a non-genomic pathway rapidly transduces estrogen signaling by involving a plasma-membrane interaction of the ER (Heldring et al. 2007; Hammes and Levin 2011). This action is mediated by GPR30, a cell-surface G protein-coupled receptor that does not act through a transcriptional mechanism (Hammes and Levin 2011). GPR30, in fact, is localized to the plasma membrane, in particular it seems that a monomeric portion of the ERa is translocated from the nucleus to the plasma membrane (Hammes and Levin 2011).

ERs are widely expressed both in reproductive and nonreproductive tissues in both men and women (Drummond and Fuller 2010; Eyster 2016). In men, ERs are expressed within the testis and through all the reproductive tract including the prostate (Rochira et al. 2016) as well as in the hearth and the cardiovascular system, brain, liver, bone, kidney, and adipose tissue (Gruber et al. 2002; Drummond and Fuller 2010; Eyster 2016).

Role of Estrogens in Men

Estrogens in men exert their direct action on several tissues and organs (Rochira et al. 2005, 2012, 2015, 2016; Table 2).

Estrogens inhibits gonadotropin secretion both at hypothalamic and pituitary level in men (Hayes et al. 2000; Raven et al. 2006; Rochira et al. 2006; Finkelstein et al. 2013) and are able to regulate male reproductive hormones; thus they ultimately regulate male reproductive function (Rochira et al. 2016; Table 2). Estrogens play also a role in the control of spermatogenesis and sperm maturation, even though the level of evidence on this estrogenic action is higher in rodents than in humans (Rochira et al. 2005, 2016; Table 2). Furthermore, estrogens are also able to influence male sexual behavior by having a positive role on sexual motivation and sexual desire (Carani et al. 1999, 2005; Rochira et al. 2005, 2012, 2016; Finkelstein et al. 2013; Table 2).

The main role of estrogens in men is within the bone tissue (Rochira et al. 2015). Estrogens, in fact, have a role both on bone maturation in peripubertal boys and in the maintenance of bone mass in adult men (Rochira et al. 2000, 2006, 2012; Vanderschueren et al. 2014; Almeida et al. 2017). At puberty, estrogen are necessary for rapid elongation of long bones followed by epiphyseal closure and growth arrest, thus allowing the attainment of harmonic skeletal proportions (between the upper and lower skeletal segments) (Zirilli et al. 2008), and the achievement of peak bone mass (Rochira et al. 2001, 2015; Table 2). Once the peak bone mass has been reached, estrogens are the main sex steroids responsible for the maintenance of bone mineral density (BMD) in men (Rochira et al. 2000, 2001, 2015; Vanderschueren et al. 2014; Almeida et al. 2017); thus, they are crucial for preventing bone loss (Finkelstein et al. 2016; Table 2).

Finally, estrogens have metabolic and vascular effects since they may positively modulate insulin sensitivity (Rochira et al. 2007) and circulating lipids (especially HDL cholesterol) (Carani et al. 1997) and be able to control fat distribution, especially at the abdominal site (Maffei et al. 2007; Finkelstein et al. 2013; Table 2). Furthermore, the lack of estrogens, but not of androgens, is involved in the appearance of vasomotor symptoms in men with severe sex steroids deficiency (Taylor et al. 2016).

Pathophysiology of Estrogen Deficiency

In men estrogen deficiency may result from the alteration of estrogen synthesis due to the genetic defect of the enzymes involved in estrogen synthesis or to an acquired reduction of the substrate (androgens) available for aromatization in men with low circulating testosterone (hypogonadism) (Rochira and Carani 2009; Trabado et al. 2011; Giton et al. 2015; Table 1, Fig. 2). Alternatively, the congenital loss-of-function of the ERs might represent a further condition of congenital loss of estrogen action (Smith et al. 1994, 2008; Table 1, Fig. 2).

Table 1 Classification of estrogen deficiency in men

Disease development	**Type of ED**	**Onset of clinical signs and symptoms**	**Grade of ED**	**Pathogenesis**
Congenital (genetic)	Primary	Prepubertal	Severe	Genetic
Acquired	Secondary to hypogonadism (relative ED)	During adulthood	Mild	Secondary to hypogonadism
				Drug induced
Estrogen deficiency in men				
Disease	**Prevalence**	**Grade**	**Causes**	
Prepubertal onset				
Congenital (genetic forms)				
Primary (classic) congenital estrogen deficiency			*Altered estrogenic pathways*	
Aromatase deficiency	Very rare	Severe	Loss-of-function mutations of the aromatase (*CYP19A1*) gene	
Estrogen resistance (one case described)	Extremely rare	Severe	Loss-of-function mutations of the estrogen receptor α gene	
Secondary congenital estrogen deficiency (relative estrogen deficiency)			*Altered estrogenic pathways*	
17 α-hydroxylase deficiency	Very rare	Mild to severe	Loss-of-function mutations of the *CYP17A1* gene	
17,20-lyase deficiency	Very rare	Mild to severe	Loss-of-function mutations of the *CYP17A1* gene	
P450 Oxydoreductase Deficiency (PORD)	Very rare	Mild to severe	Loss-of-function mutations of the *PORD* gene	
Isolated hypogonadotropic hypogonadism	Rare	Mild to severe	All genetic causes of IIH	
Acquired				
Unrecognized hypogonadism due to other diseases	Rare	Mild	Hypothalamic-pituitary diseases resulting in hypogonadism, severe primary hypogonadism (e.g., anorchia congenita)	
Adult onset				
Acquired				
Severe untreated hypogonadism	Rare to relatively frequent	Mild	Hypothalamic-pituitary diseases resulting in hypogonadism, severe primary hypogonadism; late-onset hypogonadism	

(*continued*)

Table 1 (continued)

Iatrogenic (pharmacologically induced or surgical)	Common (pharmacologically induced rare (surgical castration)	Mild to severe	Chemical castration; androgen deprivation therapy (ADT) for prostate cancer; therapy with aromatase inhibitors (e.g. breast cancer)

ED estrogen deficiency, *PORD* P450 oxidoreductase deficiency, *IIH* isolated hypogonadotropic hypogonadism, *ADT* androgen deprivation therapy

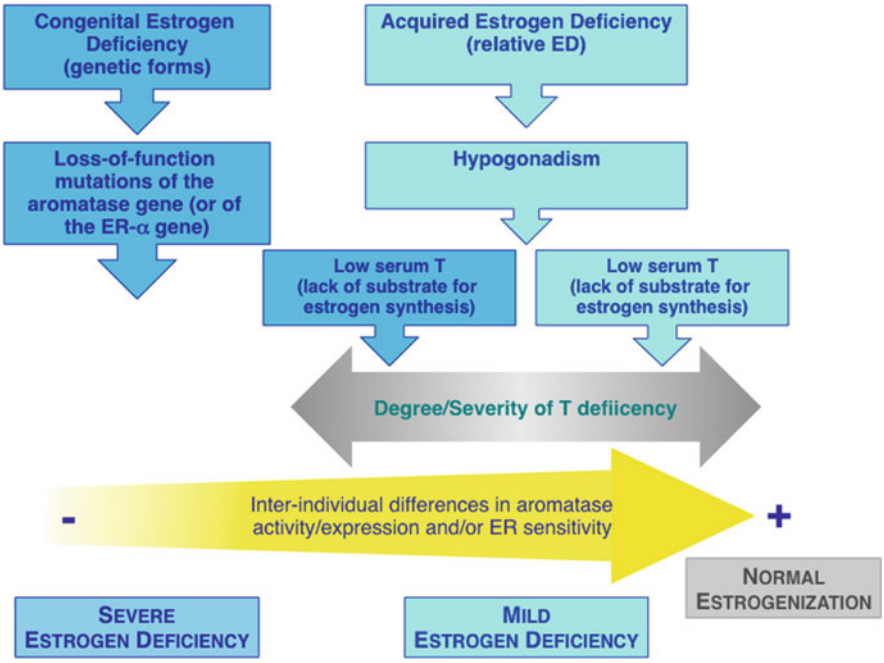

Fig. 2 Pathophysiological mechanisms of estrogen deficiency in men. *ED* estrogen deficiency, *ER* estrogen receptor, *T* testosterone

Since 1994, several cases of estrogen deficiency due to genetic defects of the ERα or of the aromatase enzyme have been described in literature (Smith et al. 1994, 2008; Rochira et al. 2002b; Zirilli et al. 2008). Furthermore, several lines of knockout mice lacking estrogen action were generated, and several studies focused on the role of estrogens in males by investigating the effects of the inhibition of estrogen synthesis in both rodents (Seralini and Moslemi 2001) and humans (de Ronde and de Jong 2011).

Thanks to the similarity between human and animal models of estrogen deficiency (Table 2), the comparison of the effects of estrogen deprivation in men with congenital estrogen deficiency with those of animal models permitted to better

Table 2 Features of estrogen deficiency in animal models and in men with congenital and acquired estrogen deficiency

Estrogen actions	Animal models of ED	Human congenital ED	Human acquired mild ED
Bone			
Reduced BMD	+++	+++	+++
Delayed epiphyseal fusion	n.a.		
Eunuchoid skeletal proportions	n.a.	+++	+
Reproductive function			
Impaired spermatogenesis	+++	+/−	n.a.
Impaired sexual behavior	+++	++ (impaired sexual desire)	+/− (impaired sexual desire)
Inhibition of gonadotropin secretion	+++	+++	+++
Glucose and lipid metabolism			
Insulin resistance	+++	+++	++ (?)
Impaired serum lipids	+++	+++	++
Adipose tissue			
Body fat accumulation	+++	+++	+++

BMD bone mineral density, *ED* estrogen deficiency, *n.a.* not applicable

clarify the clinical phenotype of estrogen deficiency, the role of estrogens in males, and species-specific estrogen actions (Faustini-Fustini et al. 1999; Grumbach and Auchus 1999; Rochira et al. 2002; Simpson et al. 2005; Jones et al. 2006; Simpson and Jones 2006).

Animal Models of Estrogen Deficiency

At the beginning, estrogen deficiency was studied in mice treated with aromatase inhibitors at dosages able to suppress quite completely estrogen production and secretion (Seralini and Moslemi 2001). These experiments investigated the effects in mice of aromatase inhibitors administered at birth or at different life periods. Once biotechnological advances made possible the genetic modification of genes involved in estrogen action, the effects of estrogen deprivation were investigated in genetically modified mouse models of estrogen deficiency.

Mice Knockout Models of Estrogen Deficiency

Several lines of mouse models lacking estrogen activity have been generated by the means of inactivation of genes encoding for estrogen receptors or involved in the synthesis of estrogens (i.e., aromatase) (Couse and Korach 1999; Hamilton et al. 2014). Four models of knockout mice are currently available: (i) the knockout of the ERα that generated the αERKO mice with nonfunctioning ERα (Lubahn et al. 1993); (ii) the knockout of the estrogen receptor beta ERβ that produced the βERKO mice

Table 3 Features that characterize the phenotype of the different knockout mice lacking estrogen action

Infertility	αERKO, αβERKO, ArKO
Impaired sexual behavior	αERKO, αβERKO, ArKO
Increased circulating gonadotropins	αERKO, αβERKO, ArKO
Increased circulating testosterone	αERKO
Increased circulating estrogens	αERKO, αβERKO,
Undetectable circulating estrogens	ArKO
Increased adiposity	ArKO
Metabolic alterations (insulin resistance, altered glucose tolerance, alteration of circulating lipids)	αERKO, αβERKO, ArKO
Impaired BMD (loss of bone mass)	αERKO, αβERKO, ArKO

with nonfunctioning ERβ (Krege et al. 1998); (iii) the knockout of both ERα and ERβ resulting in the αβERKO mice characterized by the lack of activity of both ERs (Ogawa et al. 2000); and finally (iv) the knockout of the aromatase (*CYP19*A1) gene that lead to the creation of ArKO mice, which are characterized by the lack of both circulating and locally produced estrogens (Fisher et al. 1998; Couse and Korach 1999). These models of knockout mice provided valuable information on the effects of the complete suppression of estrogen action (αβERKO and ArKO mice) as well as on the loss of action of each estrogen receptor (αERKO and βERKO mice) in males (Couse and Korach 1999; Simpson and Jones 2006). In particular, the phenotype (Table 3) arising by inactivating estrogen pathways of action in male mice allowed to better clarifying several physiological role of estrogens in rodents (Couse and Korach 1999; Simpson and Jones 2006). Furthermore, the comparison among different mouse models permitted to characterize the way in which either or both isoforms of the receptor are involved in any given action of estrogens (Heldring et al. 2007). As expected, the phenotype of male mice models leading to estrogen deficiency varies on the basis of the type of knockout (Couse and Korach 1999; Simpson et al. 2005; Table 3).

The phenotype of estrogen-deficient mouse models is similar in αERKO and ArKO male mice (Table 3), both these two mice models display altered spermatogenesis, reduced fertility, impaired sexual behavior, reduced bone mineral density, progressive increase of adiposity, and abnormal hormone serum concentrations (Rissman et al. 1997; Couse and Korach 1999; Simpson et al. 2005; Simpson and Jones 2006; Hamilton et al. 2014; Table 3). Normal to increased serum luteinizing hormone (LH) and elevated testosterone characterize the hormonal pattern of male αERKO, αβERKO, and ArKO mice; serum estradiol is increased in both αERKO and αβERKO mice (Hamilton et al. 2014) while is undetectable in ArKO mice (Fisher et al. 1998; Couse and Korach 1999; Table 3). The increase of gonadotropins is due to the lack of inhibitory feedback exerted by estrogen on the pituitary and

hypothalamus. As a consequence, the increase of LH leads to the rise of testosterone production by the testis (Couse and Korach 1999). Vice versa the negative effects of estrogen deficiency on spermatogenesis are mainly due to the lack of estrogen action (Rochira et al. 2016).

The phenotype of male βERKO mice is very close to that of wild type without particular alterations of the reproductive function and of the skeleton (Krege et al. 1998; Couse and Korach 1999; Hamilton et al. 2014).

Recently, also a knockout model of mice lacking a functioning GPR30 estrogen receptor (a transmembrane nonnuclear receptor) has become available showing a phenotype characterized by insulin resistance and altered lipid metabolism (Sharma et al. 2013).

These experimental models highlighted the role of ERα in mediating estrogen actions on male bone (Vanderschueren et al. 2014) and reproduction (spermatogenesis and male sexual behavior), αERKO adult mice being completely infertile (Rochira et al. 2005, 2016). The hormonal pattern of these mouse models of estrogen deficiency disclosed the major role of estrogens on gonadotropin feedback (Rochira et al. 2012, 2016; Table 2). Besides, models of male estrogen-deficient mice (both genetically and pharmacologically determined) are of concern since they are very close to that of humans with estrogen deficiency; this allows to extend several pathophysiological aspects to humans (Jones et al. 2006; Simpson and Jones 2006; Table 2).

Pharmacologically Induced Mouse Models of Estrogen Deficiency

The pharmacological blockade of estrogen synthesis by means of the administration of aromatase inhibitors is able to induce the same effects seen in ArKO mice in terms of bone loss (Vanderschueren et al. 1997; Eshet et al. 2004), development of infertility (Turner et al. 2000), impaired male sexual behavior (Bonsall et al. 1992; Vagell and McGinnis 1997), and hormonal pattern (Bhatnagar et al. 1992). This kind of experiment predicted about 10 years earlier several concepts on the role of estrogens in males that were subsequently revealed once estrogen deficient knockout mice were generated.

Human Estrogen Deficiency

Estrogen deficiency in men could be considered a novel nosological entity that belongs to and overlaps with several forms of male hypogonadism. At present, no specific classification of estrogen deficiency has been proposed. The disease is classified among hypogonadism or among the disorders of steroidogenesis, but a systematic clinical overview is lacking (Buvat et al. 2013; Rey et al. 2013; Huhtaniemi 2014; Dean et al. 2015). In an attempt to provide a classification of estrogen deficiency in men, the following aspects should be considered: (i) the onset of the disease (congenital/acquired); (ii) if estrogen deficiency is primarily due to alterations of estrogen pathways (primary estrogen deficiency) or if is secondary to

hypogonadism (relative estrogen deficiency); (iii) the onset of clinical manifestation; (iv) the severity of estrogen deficiency; and (v) the pathogenesis (Table 1).

Pathogenesis and Classification

Primary, classic estrogen deficiency in men is due to genetic forms that are characterized by the complete blockade of estrogen synthesis or of estrogen action (ER blockade) and lead to severe congenital estrogen deficiency without androgen insufficiency (Rochira and Carani 2009; Table 1, Fig. 2). Congenital estrogen deficiency is already present at birth without evident clinical manifestations and develops from infancy to puberty becoming completely manifest in young adults (Rochira and Carani 2009; Table 1, Fig. 2).

Secondary congenital forms of estrogen deficiency are due to genetic diseases causing androgen deficiency, which leads to relative estrogen deficiency due to reduced androgen substrate available for being transformed into estrogens (Figs. 1 and 2). In all these cases, estrogen deficiency is less severe but of clinical relevance (Table 1).

Otherwise acquired estrogen deficiency may develop at any time even though it is more frequent during adulthood (acquired hypogonadism in infancy is rare and is mainly due to hypothalamic pituitary surgery) as a consequence of hypogonadism (relative estrogen deficiency) (Trabado et al. 2011; Giton et al. 2015). In these cases, the severity of estrogen deficiency depends on that of hypogonadism (Rochira et al. 2006; Trabado et al. 2011; Giton et al. 2015; Decaroli and Rochira 2016; Table 1, Fig. 2).

Clinical Aspects of Estrogen Deficiency in Men

Congenital, Genetic Estrogen Deficiency

Pathogenesis

Primary Congenital Estrogen Deficiency

Congenital estrogen deficiency is a very rare disease listed in Orphanet. (http://www.orpha.net/consor/cgi-bin/Disease_Search.php?lng=EN&data_id=8670&Disease_Disease_Search_diseaseGroup=estrogen-deficiency&Disease_Disease_Search_diseaseType=Pat&Disease(s)/group%20of%20diseases=Aromatase-deficiency&title=Aromatase-deficiency&search=Disease_Search_Simple).

The inheritance is autosomal recessive both in case of estrogen resistance and aromatase mutations (Rochira et al. 2002; Rochira and Carani 2009).

Notwithstanding the old view considering estrogen deficiency of the fetus not compatible with life (Grumbach and Auchus 1999; Zirilli et al. 2008), several cases of men with congenital estrogen deficiency have been progressively described and contributed to really establish the role of estrogens in men throughout the whole lifetime (Rochira and Carani 2009). While only one man with estrogen resistance

due to a loss-of-function mutation of the gene encoding for the ERα is known (Smith et al. 1994, 2008), at present 14 cases of aromatase deficiency have been described in literature (Morishima et al. 1995; Carani et al. 1997; Bilezikian et al. 1998; Deladoey et al. 1999; Rochira et al. 2000; Herrmann et al. 2002, 2005; Pura et al. 2003; Bouillon et al. 2004; Mittre Herve et al. 2004; Maffei et al. 2004, 2007; Lanfranco et al. 2008; Zirilli et al. 2008; Rochira and Carani 2009; Baykan et al. 2013; Pignatti et al. 2013; Chen et al. 2015; Miedlich et al. 2016; Table 1).

Estrogen deficiency due to aromatase deficiency is the consequence of inactivating mutation of the *CYP19A1* gene encoding for the transcript of the aromatase enzyme. Several mutations have been identified and in all patients these mutations lead to a nonfunctioning aromatase protein or do not allow the transcription of the protein, which in this case is absent (Rochira and Carani 2009; Baykan et al. 2013; Pignatti et al. 2013).

Secondary Congenital Estrogen Deficiency

All the genetic diseases causing severe androgen deficiency since birth induce relative estrogen deficiency. Thus, all the diseases due to genetic alterations of steroidogenetic enzymes that result in very low circulating androgens as well all genetic diseases directly causing isolated hypogonadotropic hypogonadism might be considered as forms of congenital estrogen deficiency. Furthermore, also congenital primary hypogonadism (e.g., congenital anorchia) should be listed among congenital causes of relative estrogen deficiency (Buvat et al. 2013; Dean et al. 2015). For a comprehensive review on the classification of male hypogonadism, see the following articles deficiency (Buvat et al. 2013; Huhtaniemi 2014; Dean et al. 2015).

Different from primary congenital estrogen deficiency (which is often overlooked and undermanaged until adulthood), these diseases manifest early at birth and during infancy (ambiguous genitalia) or at puberty (e.g., isolated hypogonadotropic hypogonadism and congenital anorchia); thus the starting of adequate androgen replacement therapy at puberty avoids the appearance of most of the clinical features of congenital estrogen deficiency. If untreated or if the treatment is delayed, some clinical features like eunuchoid appearance may develop and be present in adulthood (Zirilli et al. 2008).

Clinical Phenotype

The clinical phenotype of patients with estrogen resistance is very close to that of aromatase deficiency, except for the levels of circulating hormones and the absent response to estrogen treatment in the former (Rochira et al. 2002; Bulun 2014).

The signs and symptoms of congenital estrogen deficiency are listed in Table 4 according to the time of onset of the disease.

Clinical Features in Infancy and Puberty

As the production of sex steroids is substantially silenced during infancy (Bay et al. 2004), the signs and symptoms of congenital estrogen deficiency become evident only at puberty and in young adults. Furthermore, the diagnosis is often overlooked

Table 4 Clinical features of estrogen deficiency in men according to the age of onset of the disease

Prepubertal estrogen deficiency
Delayed bone age during late adolescence
Unfused epiphyses in young male adults
Continuing linear growth during adulthood
Lack of pubertal spurt
Lack of growth arrest during late puberty
Tall stature
Eunuchoid skeletal proportions
Failure in reaching peak bone mass (osteopenia/osteoporosis)
Elevated gonadotropins
Undetectable estradiol (aromatase deficiency)
Elevated estradiol (estrogen resistance)
Not always present:
Genu valgum
Scoliosis
Cryptorchidism
Secondary to congenital or acquired hypogonadism:
Abnormalities of external genitalia
Delayed puberty
Postpubertal estrogen deficiency
Osteoporosis/osteopenia
Diffuse bone pain
Reduction of sexual desire
Early-onset metabolic syndrome
Fat redistribution and accumulation (abdominal adiposity)
Dyslipidemia
Acanthosis nigricans
Nonalcoholic fatty liver disease
Acquired (relative estrogen deficiency)
Signs and symptoms of hypogonadism

for a variable period and is usually reached during adulthood (mean age of described cases is about 30 years) (Rochira and Carani 2009).

During infancy, the disease could be suspected only if the anamnestic information regarding mother's virilization during the last trimester of pregnancy is available or in case of affected relatives (Faustini-Fustini et al. 1999; Grumbach and Auchus 1999; Rochira and Carani 2009). History of mother's virilization during pregnancy is mandatory to prompt prenatal diagnosis by means of amniocentesis (Rochira and Carani 2009; Bulun 2014; Morel et al. 2016).

At puberty, clinical evidence for the disease remains, however, difficult to unmask due to the fact that the course of the disease is characterized by few symptoms (Rochira and Carani 2009). The most evident sign of the disease at puberty seems to be the lack of pubertal growth spurt with the height continuing

to increase linearly as in children (Lee and Witchel 1997; Faustini-Fustini et al. 1999; Grumbach and Auchus 1999; Rochira et al. 2001; Table 4). Accordingly, the pubertal growth spurt seems to be completely lacking in aromatase deficiency (Lee and Witchel 1997; Faustini-Fustini et al. 1999; Grumbach and Auchus 1999; Rochira et al. 2001). In confirmation of this, pubertal spurt was absent before starting the estradiol replacement treatment at the age of 17 (Bouillon et al. 2004) in the child with early diagnosis (Deladoey et al. 1999).

In clinical practice, however, the lack of pubertal growth spurt can be easily overlooked, since it is a very difficult sign to spot in a growing boy unless a clinician keeps the height constantly monitored on the growth chart. If we additionally consider the interindividual differences in the onset and progression of the pubertal process, aromatase deficiency appears unlikely to be suspected at puberty. Furthermore, the other two associated signs (eunuchoid body proportions of the skeleton and delayed bone maturation) that are slowly developing at that time are also very difficult to detect at an early stage of the disease. For all these reasons, estrogen deficiency is not diagnosed at puberty as well as in young adults, while it is more likely to be detected in adulthood (Rochira and Carani 2009). At present, only one case was diagnosed at birth thanks to the already available diagnosis in a relative (Deladoey et al. 1999; Bouillon et al. 2004).

In case of secondary congenital estrogen deficiency, the main clinical features are related to androgen deficiency ranging from hypogenitalism at birth to overt ambiguous genitalia in case of defects of the steroidogenesis (Fluck and Pandey 2014; Auchus 2017). In case of congenital isolated hypogonadotropic hypogonadism usually the delay of the onset of puberty prompts further clinical investigations and allows reaching the diagnosis in pubertal boys (Bonomi et al. 2012).

Clinical Features in Adulthood

Signs and symptoms of congenital estrogen deficiency remain challenging also in adulthood (Rochira and Carani 2009). Patient's stature does not stop but continues to grow at such a small rate that it does not become evident as a pathological condition. As a consequence, also the eunuchoid body proportions develop very slowly and are not easily recognized as a clinical problem. The occasional finding of unfused epiphyses at X-ray examinations in an adult man may prompt the diagnosis (Rochira and Carani 2009).

For all these reasons, the congenital estrogen deficiency remains largely undiagnosed until adulthood and its diagnosis delayed even during adulthood when signs and symptoms become more evident but are not recognized (Rochira and Carani 2009).

The hormonal phenotype of patients with estrogen resistance is characterized by elevated serum levels of estradiol, luteinizing hormone (LH), follicle-stimulating hormone (FSH), and normal serum testosterone (Smith et al. 1994, 2008; Rochira et al. 2002). Vice versa, the hormonal phenotype of aromatase-deficient men is characterized by undetectable serum estradiol, elevated gonadotropins, especially FSH, and by variable serum testosterone, ranging from values slightly below the

lower end of the normal range to values slightly above the upper limit of the normal range, but being normal in most of the cases (Rochira and Carani 2009).

The clinical phenotype of aromatase-deficient men may show interindividual differences (Rochira et al. 2002), but the following clinical features are always present in adults: tall stature, diffuse bone pain, continuing, linear growth in adulthood (if untreated), delayed bone maturation and unfused epiphyses, eunuchoid proportions of skeletal segments, genu valgum, tendency to fat abdominal accumulation, osteopenia, or osteoporosis (Rochira et al. 2002; Zirilli et al. 2008; Rochira and Carani 2009). Interindividual differences in the presentation of the disease in adulthood involve the following signs and symptoms: metabolic alterations (insulin resistance, *acanthosis nigricans*, metabolic syndrome, nonalcoholic fatty liver, dyslipidemia), reduced fertility (oligozoospermia), macroorchidism, and scoliosis (Rochira and Carani 2009).

Nothing is known about the clinical phenotype of partial aromatase deficiency in men since no cases have been described. In female, partial aromatase deficiency leads of a very mild phenotype (Lin et al. 2007; Pepe et al. 2007; Gagliardi et al. 2014).

When congenital estrogen deficiency is secondary to hypogonadism, defects of external genitalia and delayed puberty are present (Table 4).

Diagnosis

The diagnosis of primary, congenital estrogen deficiency in men is challenging and is almost always suspected in adulthood. The presence of affected relatives and/or of a history of mother's progressive, transient virilization during the last trimester (that spontaneously resolves after delivery) may prompt earlier the diagnosis (Morishima et al. 1995; Grumbach and Auchus 1999; Rochira and Carani 2009; Bulun 2014). Mother's virilization is characterized by severe hirsutism due to the androgen precursors that accumulate as a consequence of the blockade of placental aromatase and pass into mother's blood circulation (Morishima et al. 1995; Faustini-Fustini et al. 1999; Grumbach and Auchus 1999). Conversely, the disease is rarely overlooked in females since it is easy to diagnose at birth or at puberty thanks to the presence of ambiguous genitalia and primary amenorrhea, respectively (Belgorosky et al. 2009).

The main aspects that should be checked at interview and physical examination for the diagnosis of congenital estrogen deficiency are listed in Table 5.

In particular, the presence of relatives with congenital estrogen deficiency and the consanguinity of parents could orient toward a genetic disease (Rochira and Carani 2009). Physical examination should focus on patient's height (compared to the patient's target) and further, unexpected increase in height during adulthood, the presence/absence of the growth spurt (if past data are available), and on the skeletal proportions (Rochira and Carani 2009; Table 5). Usually, pubertal development progresses in a normal fashion and virilization is normal, while the testes are rarely enlarged (normal in most of the cases) or undescended (Rochira and Carani 2009; Rochira et al. 2016; Table 5).

Table 5 Clinical aspects to be considered during the visit for the diagnosis of congenital estrogen deficiency

Specific issues to address during the clinical interview
Information from relatives
Parents' consanguinity
Parents' height for the estimation of target stature
Maternal virilization during pregnancy
History of ambiguous genitalia and/or delayed puberty in sisters
Patient's information
Weight and length at birth
Estimate of his target height
Patient's early growth
Patient's pubertal development
History of cryptorchidism
Presence of offspring
Patient's sexual behavior (sexual identity, sexual orientation, sexual activity)
Specific issues to address during the physical examination
Anthropometric parameters
Height
Weight
BMI
Upper and lower skeleton segments lengths
Lower segment of the skeleton length
Arm span length
Waist and hip circumferences
Other clinical examinations
Degree of virilization
Testicular volume
Testes localization
Penis size
Blood pressure
Check for skeletal deformations (e.g. scoliosis or *genu valgum*)
Check for surrogates of insulin resistance (*acanthosis nigricans* and skin tags)
Clinical examinations
Hormonal assessment
Serum estradiol
Serum testosterone
Serum LH
Serum FSH
Other serum androgens (not mandatory)
IGF-1, SHBG
Biochemical analyses
Serum fasting glucose and insulin
Serum lipids
Liver function

(*continued*)

Table 5 (continued)

	Markers of bone turnover (useful for monitoring therapy)
	Sperm analysis
Radiological examinations	
	X-ray film of hand and wrist
	BMD measurement by DEXA at lumbar and femoral site
	X-ray film of the spine in case of back pain
	Liver ultrasonography
Genetic analysis	
	Aromatase or ERα gene sequencing (depending on clinical data)

LH luteinizing hormone, *FSH* follicle-stimulating hormone

Among hormonal examinations, the measurements of serum gonadotropins, estradiol, and testosterone are mandatory, while adrenal androgens are not strictly necessary (Table 5; Rochira and Carani 2009).

The diagnosis should exclude other congenital, genetic diseases that cause severe hypogonadism and/or abnormal steroidogenesis (Table 1; Rochira and Carani 2009). The finding of very low serum testosterone in the presence of low (but not undetectable serum estradiol) allows differentiating congenital estrogen deficiency from these forms (Table 5; Rochira and Carani 2009). Furthermore, these conditions are associated to various degrees of undervirilized genitalia, which are not present in congenital estrogen deficiency (Rochira and Carani 2009; Fluck and Pandey 2014; Auchus 2017; Table 4). Among congenital primary estrogen deficiency, high versus undetectable serum estradiol helps differentiating estrogen resistance from aromatase deficiency (Table 4; Rochira and Carani 2009). For this reason, the hormonal examinations together with the estimation of bone age should be considered as the first step in the diagnosis of congenital estrogen deficiency (Rochira and Carani 2009; Table 5).

Other biochemical analyses should be addressed to obtain information on the metabolic alterations and bone health status when the clinical suspicion of primary congenital estrogen deficiency is confirmed by hormonal analyses and bone age (Table 5). Accordingly, some comorbidities such as insulin resistance, dyslipidemia, abdominal obesity, fatty liver disease, and metabolic syndrome are often associated with the congenital lack of estrogen action and require specific diagnostic approaches (Rochira et al. 2002; Maffei et al. 2004, 2007; Herrmann et al. 2005; Rochira et al. 2007; Zirilli et al. 2008; Table 5).

Even in adulthood, the strength of evidence of a diagnosis based solely on clinical data (signs, symptoms, hormonal and radiological outcomes) (Table 4) is poor due to the rarity of the disease. The limits of estradiol assay in clinical practice complicate the diagnostic issue (see paragraph below for further details). Thus, the genetic analysis is strongly suggested in order to further substantiate the clinical suspect (Table 5). Accordingly, sequencing of the aromatase gene proved effective in revealing gene defects (point mutations and/or base pair deletions) in all the patients with aromatase deficiency described so far (Smith et al. 1994, 2008; Morishima et al. 1995;

Carani et al. 1997; Bilezikian et al. 1998; Deladoey et al. 1999; Rochira et al. 2000; Herrmann et al. 2002; Pura et al. 2003; Bouillon et al. 2004; Maffei et al. 2004; Mittre Herve et al. 2004; Herrmann et al. 2005; Lanfranco et al. 2008; Zirilli et al. 2008; Rochira and Carani 2009; Baykan et al. 2013; Pignatti et al. 2013; Chen et al. 2015; Miedlich et al. 2016).

The genetic analysis is useful to solve possible doubts raised by unclear clinical pattern or by the limits of estradiol assays (which are not accurate within the male range) (Table 5). Furthermore, the genetic analysis is mandatory to differentiate congenital estrogen deficiency from other forms of rare genetic alterations of steroidogenesis that share both estrogen deficiency and some clinical features (Rochira and Carani 2009; Tables 1 and 4).

Fertility should be investigated by means of sperm analysis, while a detailed sexual interview should investigate sexual health (Carani et al. 1999, 2005; Rochira and Carani 2009; Table 5).

Bone status should be studied by means of dual energy X-ray absorptiometry (DEXA) and markers of bone turnover at baseline; this work-up allows the subsequent monitoring of the efficacy of the treatment on bone (Rochira et al. 2000, 2007, 2015; Zirilli et al. 2009; Table 5).

Finally, it should be remarked that also the pituitary function, especially growth hormone secretion, may be impaired in men with congenital estrogen deficiency (Rochira et al. 2002, 2010; Table 5).

In daily clinical practice, however, the likelihood to visit a patient with clinical features suggestive for congenital estrogen deficiency is very low, and it is highly probable that most endocrinologists will never see a patient with congenital estrogen deficiency during their career due to the rarity of these diseases (Rochira and Carani 2009).

Therapy

Treatment of congenital estrogen deficiency is based on the administration of estrogens and is effective only in men with aromatase deficiency (Carani et al. 1997; Bilezikian et al. 1998; Rochira and Carani 2009) while does not have any effect in men with estrogen resistance (Smith et al. 1994).

In adult aromatase-deficient men high starting dose of estrogens (e.g., 25–50 μg of estradiol per day) are needed for a short period (about 6–9 months) soon after the disease has been diagnosed in order to quickly complete bone maturation, fuse epiphyses, reach growth arrest and final height and induce peak bone mass (Carani et al. 1997; Bilezikian et al. 1998; Rochira et al. 2000; Balestrieri et al. 2001; Rochira and Carani 2009). This dosage is effective in obtaining serum levels of estradiol ranging from the upper limit of the normal range and values that are slightly above the upper limit of the normal range (Rochira et al. 2000, 2001; Balestrieri et al. 2001). Once the epiphyses close and the vertical growth cease, the dosage should be reduced (about to 25 μg of estradiol per day) to reach more physiological levels of circulating estradiol (Balestrieri et al. 2001; Rochira and Carani 2009). This dosage should be considered for lifelong replacement therapy in order to ensure bone mass maintenance

and a healthy metabolic status (glucose, lipid, and liver metabolism) (Balestrieri et al. 2001; Rochira and Carani 2009). In adulthood, however, estrogen treatment is ineffective in restoring normal skeletal proportions and in improving sperm count and quality and has poor effect on fat redistribution (Rochira and Carani 2009).

If the disease is diagnosed before puberty, as desirable, estrogen treatment should be started at a very low dosage starting from the onset of puberty, and the dosage should be titrated by increasing the amount of exogenous estrogens according to the progression of puberty (Bouillon et al. 2004; Rochira and Carani 2009). This treatment is able to mimic the physiological changes in circulating estrogens occurring throughout puberty and to avoid the delay of bone maturation and the development of unbalanced skeletal proportions and to reach normal peak bone mass (Bouillon et al. 2004; Zirilli et al. 2008).

The estrogen formulation of choice for replacement therapy may change according to the physician's attitude and experience (all are effective), but the administration of estradiol should be preferred since it is measurable in serum and allows monitoring changes in circulating estradiol levels obtained after dose adjustments (Rochira et al. 2000), the latter being not possible with other formulations (e.g., conjugated estrogens) (Balestrieri et al. 2001; Rochira and Carani 2009). As far as safety is concerned, no adverse effects have been reported in aromatase-deficient men treated with estrogen replacement treatment (Rochira and Carani 2009).

As estrogen treatment has positive effects on bone mass in these men, they should be supplemented with calcium and vitamin D in order to better support the anabolic effects of estrogens on bone (Rochira and Carani 2009).

Adequate follow-up should be addressed to verify the completion of bone maturation (by X-ray films of hand and wrist), to ensure optimal estrogen dosage (by monitoring hormonal analyses, especially serum LH, FSH and estradiol), to monitor bone mass (by DEXA), and to check glucose and lipid metabolism (by monitoring metabolic parameters), especially if they were altered at baseline (Rochira and Carani 2009).

Estrogen treatment of the unique patient with estrogen deficiency was ineffective in improving bone mineral density, bone maturation, and other clinical features of the disease (Smith et al. 1994). Theoretically, the further increase of circulating estrogens might be a possible strategy to overcome the blockade of the ER in cases of partial estrogen resistance, the latter, however, have never been described.

Acquired Estrogen Deficiency

Acquired estrogen deficiency in men usually occurs during adulthood as a consequence of mild to severe hypogonadism independently from the cause of testosterone deficiency (Table 1, Fig. 2). Acquired forms of estrogen deficiency rarely develop in infancy or at puberty, in these cases they are mainly due to pituitary disorders or surgery (e.g., craniopharyngioma) or primary (hypergonadotropic) hypogonadism (Table 1).

Estrogen deficiency occurring during adulthood is always the consequence of reduced circulating estrogens secondary to impaired secretion of androgens (Fig. 2); thus, they should be considered as conditions of relative estrogen deficiency (Trabado et al. 2011; Giton et al. 2015; Table 1, Fig. 2).

Pathogenesis of Acquired Estrogen Deficiency

The most common condition causing relative estrogen deficiency seems to be aging, since serum estradiol decreases in parallel with testosterone with advancing age (Decaroli and Rochira 2016), as documented by several longitudinal studies in older men (Feldman et al. 2002; Wu et al. 2010; Huhtaniemi et al. 2012). Other main diseases that are strictly related to adult-onset testosterone deficiency are poor general health and obesity (Wu et al. 2010; Huhtaniemi 2014; Corona et al. 2015, 2016; Decaroli and Rochira 2016).

Diseases Causing Hypogonadism and Relative Estrogen Deficiency

All acquired hypothalamic-pituitary diseases and testicular disorders causing hypogonadism might cause concomitant estrogen deficiency (Buvat et al. 2013; Huhtaniemi 2014; Dean et al. 2015). Furthermore, all the other clinical conditions that are associated to hypogonadism should be considered (Buvat et al. 2013; Huhtaniemi 2014; Dean et al. 2015). For a comprehensive review on the classification of male hypogonadism, see the following articles deficiency (Buvat et al. 2013; Huhtaniemi 2014; Dean et al. 2015).

Iatrogenic Estrogen Deficiency

Estrogen deficiency might also be the consequence of therapies that are able to reduce both circulating androgens and estrogens or estrogens alone.

Androgen deprivation therapy for prostate cancer results in profound fall of serum testosterone and estrogens (Guise et al. 2007; Freedland et al. 2009). Both surgical and chemical castration are able to suppress circulating sex steroids of more than 90% with respect to baseline levels (Nishii et al. 2012). Thus, severe hypogonadism results from the administration of gonadotropin releasing hormone (GnRH) agonists and antagonists as well as from surgical castration (orchiectomy) (Guise et al. 2007; Freedland et al. 2009). Independently from the type of androgen deprivation therapy, estrogen deficiency is always severe consisting with serum estradiol levels below 5 pg/mL (Nishii et al. 2012) and is responsible for bone loss, osteoporosis, and a dramatic increase of fracture risk (Shahinian et al. 2005). In these patients, estrogen deficiency leads also to lipid alterations (Freedland et al. 2009) and hot flashes (Freedland et al. 2009), the latter being mainly due to estrogen deficiency rather than to low serum testosterone (Taylor et al. 2016).

Treatment of male breast cancer includes the use of aromatase inhibitors that are effective in reducing serum estradiol but are not able to completely suppress estrogens and to induce severe estrogen deficiency (Kuba et al. 2016). Accordingly, serum estradiol in men with breast cancer treated with aromatase inhibitors is reduced to values slightly below the normal male range (Kuba et al. 2016). Even

in these patients, pharmacologically induced estrogen deficiency increases the risk of osteoporosis and of osteoporotic fractures (Saad et al. 2008).

Aromatase inhibitors are also used to increase adult height in boys with idiopathic short stature, precocious puberty, and constitutional delay of puberty (de Ronde and de Jong 2011). The strategy to increase final height is based on the induction of estrogen deficiency in order to delay epiphyseal closure and extend the period of growth (Wit et al. 2011), but this therapeutic approach might be dangerous in terms of bone health since bone deformities have been described to develop in these young patients (Hero et al. 2010).

Clinical Significance of Relative Estrogen Deficiency

Estrogen deficiency becomes harmful for male health only when serum estradiol falls significantly (Mellstrom et al. 2008; Finkelstein et al. 2013; Rochira et al. 2015). In particular, detrimental effects of estrogen deficiency on bone develop only when serum estradiol falls below a threshold that has been settled around 20 pg/mL (Mellstrom et al. 2008; Rochira et al. 2015) and the same seems to happen for other male physiological functions such as sexual behavior and tendency to fat mass increase (Finkelstein et al. 2013). Thus, in the presence of low serum testosterone, some health problems appear and/or worsen only when also relative estrogen deficiency is present (Decaroli and Rochira 2016).

Several studies demonstrated that not only bone health but also male sexual behavior, fat redistribution, and accumulation as well as glucose and lipid metabolism depend on estrogen deficiency (Rochira et al. 2016). How deep should hypogonadism be, in terms of low serum testosterone, to induce clinically relevant estrogen deficiency is difficult to define due also to the probable existence of interindividual differences in aromatase activity and expression and/or ERs sensitivity (Fig. 2; Rochira et al. 2006, 2015; Vottero et al. 2006). It is clear that hypogonadal men who are protected by higher circulating estrogens might be at lower risk of developing signs and symptoms specifically related to estrogen deficiency (e.g., bone loss and osteoporosis) (Rochira et al. 2006; Aguirre et al. 2015). At present, however, this is a poorly investigated issue. Only two studies speak in favor of the existence of interindividual differences in aromatase function that may exacerbate or protect from estrogen deficiency depending on decreased or increased interindividual aromatase function (Fig. 2). The first one postulates that different aromatase activity may lead to different estrogenization starting from a given serum testosterone (Vottero et al. 2006); the second shows that high aromatase activity exerts protective effect on bone loss in hypogonadal men (Aguirre et al. 2015). Further clinical data come from studies focusing on the relationship between the severity of hypogonadism and that of estrogen deficiency, another important issue in determining the degree of hypoestrogenism (Fig. 2). Hence, the study of serum estradiol in hypogonadal men with or without testosterone replacement therapy indicates that severe estrogen deficiency (<10 pg/mL) is constantly present in untreated hypogonadal men with severe hypogonadism (serum testosterone below 50 ng/dL) (Trabado et al. 2011). Furthermore, adult men with untreated isolated hypogonadotropic hypogonadism have severe estrogen deficiency, but residual very

low circulating estrogens remain detectable by liquid chromatography-tandem mass spectrometry (LC-MS/MS), while in men with untreated panhypopituitarism estrogen deficiency is associated with undetectable serum estrogens due to concomitant absence of androgens not of testicular origin (Giton et al. 2015).

The knowledge of relative estrogen deficiency in adult men remains still poor, and further researches are needed. At present the best evidence comes from the group of Joel S. Finkelstein who designed an experimental placebo-controlled study on healthy men who underwent pharmacologically induced hypogonadism by means of goserelin acetate administration. The addition of various doses of testosterone allowed obtaining five different groups of patients differing in serum testosterone. Further administration of aromatase inhibitors allowed obtaining various degree of estrogen deficiency and to unravel the role of both sex steroids (androgens and estrogen) on several male functions (Finkelstein et al. 2013, 2016; Taylor et al. 2016). This experimental setting demonstrated that sexual function needs both estrogens and androgens to be fully normal that visceral and subcutaneous fat accumulation is prompted by estrogen deficiency (Finkelstein et al. 2016) and that bone loss (Finkelstein et al. 2016) and vasomotor symptoms (Taylor et al. 2016) are mainly due to estrogen deficiency.

Relative Estrogen Deficiency: Clinical Aspects

As relative estrogen deficiency is always a consequence of hypogonadism (with the exception of estrogen deficiency induced by aromatase inhibitors), the prevailing clinical features are those of hypogonadism (Buvat et al. 2013; Huhtaniemi 2014; Dean et al. 2015). However, among signs and symptoms of hypogonadism (i.e., reduced muscle strength, fatigue, impaired well-being, reduced energy and motivation, mood disturbances, reduced sexual desire, impaired sexual function, hot flashes, bone loss, fat accumulation) (Buvat et al. 2013; Huhtaniemi 2014; Dean et al. 2015), unraveling those mainly ascribable to estrogen deficiency remains challenging. Bone loss, osteoporosis/osteopenia, hot flashes, fat accumulation, and redistribution depend mainly on estrogen deficiency (Finkelstein et al. 2013, 2016; Taylor et al. 2016). Other clinical manifestations such as alterations of sexual desire and function probably are the consequence of both estrogen and androgen deficiency.

To Measure or Not Estrogens in Men? The Clinical Dilemma

The diagnosis of relative estrogen deficiency is usually made within the context of the clinical work-up programmed to diagnose hypogonadism. With this in view, the diagnosis of relative estrogen deficiency should be considered as the refining of the diagnosis of hypogonadism. The diagnosis of relative estrogen deficiency is mainly based on the finding of serum estradiol below the normal male range. However, serum estradiol is not always assayed in men in the *real life* of clinical practice, either in case of suspected or of already documented hypogonadism. Current guidelines or expert committee opinions on the clinical management of hypogonadism, in fact, do not recommend the measurement of circulating estrogens within the clinical work-up for the diagnosis and follow-up of male hypogonadism (Petak et al. 2002;

Wang et al. 2009; Bhasin et al. 2010; Buvat et al. 2013; Huhtaniemi 2014; Dean et al. 2015).

Theoretically, the measurement of serum estradiol is needed in order to establish if a man with hypogonadism has concomitant relative estrogen deficiency or not and/or if replacement androgen treatment ensures normal estrogenization. The information on the presence of estrogen deficiency is useful to stratify patients risks related to hypoestrogenism (e.g., osteoporosis) (Rochira et al. 2006; Aguirre et al. 2015) and to orient about the need of further clinical examinations (e.g., DEXA). In clinical practice, however, the measurement of estradiol in men raises more doubts than answers due to pitfalls related to the measurement of estradiol in the low male normal range (Santen et al. 2015). At present only the American Association Clinical Endocrinologists suggests that androgen treatment of hypogonadal men should keep also estrogens within the male physiological range but does not provide advice on how to approach this matter from a clinical point of view (Petak et al. 2002).

Outside the context of congenital, genetic forms of estrogen deficiency, the main problem concerning estrogen deficiency lies in the poor accuracy of commercially available assays when measuring circulating estrogens within the low male range (Handelsman et al. 2014; Taylor et al. 2015). Accordingly, commercially available kits, which remain the most used method for assaying estrogens in the clinic, are not reliable in men due to their poor accuracy, insufficient sensitivity, and lack of reproducibility (Rosner et al. 2013; Demers et al. 2015). Furthermore, immunoassays provide less reproducible results compared to LC-MS/MS (Handelsman et al. 2014). Vice versa LC-MS/MS is a valuable method for measuring even very low amounts of circulating estrogens, but it is mainly used for research purposes (Huhtaniemi et al. 2012; Rosner et al. 2013). At present, the gold standard LC-MS/MS is becoming more and more available in the clinical setting (Simoni et al. 2012; Santen et al. 2015,; Simpson and Santen 2015), but at the moment, it is rarely used in clinical laboratories (Santen et al. 2015; Simpson and Santen 2015). Besides, how to interpret the results obtained by LC-MS/MS in the absence of standardized reference male ranges remains challenging (Demers et al. 2015; Santen et al. 2015; Simpson and Santen 2015). Serum estradiol levels obtained by LC-MS/MS in men, in fact, remain not validated for clinical use (Demers et al. 2015). In addition, the validation of estrogen measurement by LC-MS/MS is not so easy due to technical issues with sex steroids. Thus, the measurement of serum estrogens in men is considered unessential in clinical practice at the moment (Demers et al. 2015).

In any case, clinical laboratories offer estrogen measurement for males in the daily *real life*, independently from the type of method used and clinicians ask for estrogen measurement. Notwithstanding the poor accuracy and the advice of guidelines and experts, clinicians probably consider the measurement of serum estrogens to be of some use for their practice. Even though imprecise the information on circulating estrogens could be useful to have an idea on patient's estrogen status, on the risks related to estrogen deficiency, and on the effect of testosterone treatment on serum estrogens. Because immunoassays tend to overestimate low serum estradiol

levels, they fail in detecting all patients with relative estrogen deficiency, but when a value of serum estradiol below the normal range is obtained, the diagnosis of estrogen deficiency is more reliable. Furthermore, considering that commercially available immunoassay are more accurate for detecting elevated estradiol levels in men (Huhtaniemi et al. 2012), serum estradiol measurement may be useful to avoid overtreatment in hypogonadal men under testosterone replacement therapy.

Conclusions

While the role of estrogens in men and the harmful potential of estrogen deficiency are well known, the necessity to investigate estrogen deficiency from a clinical point of view, outside the context of rare congenital, genetic forms remains to be determined. Furthermore, the information on the presence/absence of estrogen deficiency is of little help for the patient, since the only therapeutic strategy available is that of using testosterone (or other aromatizable androgens). Accordingly, estrogen therapy in men with hypogonadism increases estrogen levels but inhibits gonadotropins and further decreases endogenous testosterone production.

Information on male estrogen status will potentially be useful in the future if developed new drugs are able to selectively stimulate aromatase, increase endogenous estrogens, and help in balancing estradiol end testosterone.

At present, adjunctive information on the levels of circulating estrogens and of estrogen to testosterone ratio is of relevance for setting patient's risk of developing features strictly related to estrogen deficiency (e.g., osteoporosis) as well as to monitor the effects of testosterone on estrogens during the follow-up of hypogonadal men under testosterone replacement therapy (Matsumoto 2013).

Finally, some unresolved issues still remain. Among them, we do not know the impact of estrogen deficiency in boys before infancy. Even though circulating serum sex steroids are very low during infancy, low amounts of both androgens and estrogens may be detected by using ultrasensitive assays (Bay et al. 2004), the role of which remains to be determined. Thus, the need of estrogen treatment of a child with documented congenital estrogen deficiency is unknown. Besides, the contribution of estrogen deficiency on morbidity and mortality in men needs to be determined since the studies that consider androgens and estrogens separately are scanty (Jankowska et al. 2009; Tivesten et al. 2009; Hsu et al. 2016).

Key Points

Estrogen deficiency in men is a heterogeneous clinical condition.
A systematic classification of estrogen deficiency is lacking.
Congenital estrogen deficiency is an overlooked and undermanaged, rare disease.
Relative estrogen deficiency is common among hypogonadal men.
Poor accuracy of serum estrogen assays undermines the clinical approach to estrogen deficiency in adult men.

Summary

Genetic estrogen deficiency in men is rare while relative estrogen deficiency due to male hypogonadism is a common clinical condition. In clinical practice, however, estrogen deficiency is poorly considered due to its rarity (genetic forms) or to the fact that physicians focus their attention mainly on hypogonadism, the primary disease, in the case of relative estrogen deficiency. For these reasons, the promotion of the know-how concerning the physiological and pathological role of estrogens and their insufficiency in men is important to improve, among clinicians, the awareness on this pathological condition.

Cross-References

- ▶ Anabolic and Metabolic Effects of Testosterone and Other Androgens: Direct Effects and Role of Testosterone Metabolic Products
- ▶ Gonadotropins
- ▶ Late-Onset Hypogonadism
- ▶ Testicular Steroidogenesis

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