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Role of estrogen on bone in the human male: insights from the natural models of congenital estrogen deficiency

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Abstract

The reports of congenital estrogen deficiency — notably, estrogen resistance and aromatase deficiency — have completely changed our knowledge on the role of estrogen on bone in males. Particularly, the bone changes at puberty, which were classically considered androgen-dependent, are now considered to be induced at least in part by estrogen action. Clinical cases of congenital estrogen deficiency have clearly demonstrated that the role of estrogens in epiphyseal closure, skeletal proportions and bone mineralization is crucial not only in women but also in men. In addition progress have been made in the treatment of such a rare disease even though further studies are needed to a definitive understanding of this issue.

Keywords: Estrogen deficiency; Bone maturation; Bone mineralization; Eunuchoid skeleton; Male osteoporosis; Estrogen replacement therapy

1. Introduction

Recently, remarkable progress has been made in our understanding of the role of sex steroids on growth and skeletal maturation.

In humans, male testosterone and estradiol are the main circulating sex steroidal hormones acting on bone tissue. Testosterone is produced by the Leydig cells in the testis, while estrogen comes from the aromatization of androgens by means of the action of the aromatase cytochrome-P450 enzyme.

The biological effects of estrogens are mediated by binding with the nuclear estrogen-receptors (ER) and to date, two different estrogen-receptors, ER-α and ER-β have been characterized (Enmark and Gustafsson, 1999). Besides, a non-genomic pathway of estrogen action has recently been documented, which probably involves a plasma-membrane ER interaction (Levin, 1999).

The production of transgenic mice which are either estrogen resistant (Couse and Korach, 1999) or do not produce estrogens (Fisher et al., 1998), together with the discovery of naturally occurring inactivating mutations of both the estrogen receptor (ER) and the aromatase gene in humans, opened new prospects for a better understanding of estrogen physiology in the human male (Faustini-Fustini et al., 1999; Grumbach and Auchus, 1999).

2. Estrogen and bone maturation

Three well-documented cases of congenital estrogen deficiency in adult men have been reported in the literature (Faustini-Fustini et al., 1999); all but one were the consequence of an inactivating mutation of the aromatase gene (aromatase deficiency) (Morishima et al., 1995; Carani et al. 1997), while the other one still remains the unique case due to an inactivating mutation of the ER-α gene (estrogen resistance) (Smith et al., 1994). The patient's phenotype is characterized by tall stature, a history of continuing linear growth into adulthood, unfused epiphyses, delayed bone age, osteoporosis, eunuchoid proportions of the skeleton, and progressive genu valgum. No skeletal abnormalities

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were recently reported in a male infant affected by aromatase deficiency (Deladoëy et al., 1999).

3. Estrogen and pubertal growth spurt

By history, no abnormalities of both early growth and pubertal development were reported in congenital estrogen-deficient men. It has been supposed that pubertal growth spurt does not occur in men with congenital estrogen deficiency, thus remaining longitudinal growth (a linear process) during puberty and adulthood as it happens in the prepubertal period. With this in mind, the male pubertal growth spurt could be considered an estrogen-dependent process (Grumbach and Auchus, 1999), but uncertainties still remain, as data on pubertal growth of estrogen-deficient adult men are not available in detail (growth curves are incomplete) (Faustini-Fustini et al., 1999). The discovery of novel mutations in male infants (Deladoëy et al., 1999) together with the chance of monitoring their height continuously from childhood to adulthood will probably disclose the natural history of estrogen deficiency, improving our knowledge of this issue.

In boys with idiopathic delay of puberty, very low doses of estradiol treatment, which correspond to estradiol levels of approximately 4 pg/ml, a value detectable only by using an ultrasensitive cell bioassay, resulted in a maximal ulnar growth, having at higher doses, a minor or even an inhibitory effect on epiphyseal growth (Caruso-Nicoletti et al., 1985; Cutler Jr., 1997). By analyzing a model of pure unopposed estrogen action, as it is complete androgen insensitivity syndrome (CAIS), a spontaneous pubertal growth and a complete skeletal maturation occur, despite the lack of androgen action (Zachmann et al., 1986). These two models clearly suggest a key role for estrogens in pubertal skeletal growth.

4. Epiphyseal closure and growth arrest

Estrogen treatment leads to epiphyseal closure in adult men with aromatase deficiency (Carani et al., 1997; Bilezikian et al., 1998), being inefficacious in the estrogen-resistant man (Smith et al., 1994). By contrast, testosterone treatment was not able to promote skeletal maturation in one of the two aromatase-deficient adult men (Carani et al., 1997). These findings clearly demonstrate that, even in the male, epiphyseal closure does not develop without estrogens and that androgens alone are not able to complete bone maturation.

Estrogen role on skeletal maturation becomes clearer if we consider that accelerated growth, advanced bone age, and probably a short final stature constitute the clinical features of the aromatase excess syndrome in both sexes (Stratakis et al., 1998), an opposite phenotype of that of aromatase deficiency.

5. Estrogen and skeletal proportions

A further fascinating question involves the role of sexual steroids in affecting the normal body proportions of the skeleton. Traditionally, it was thought that eunuchoid body habitus, i.e. a preferential growth of the arms and legs compared with the spine — comes from an impairment of androgen action. The finding of eunuchoid body proportions in men with congenital estrogen deficiency having normal or elevated testosterone serum levels implies that estrogens are involved in the establishment of the proportions of the growing skeleton. Accordingly, subjects with CAIS show normal skeletal proportions despite the absence of androgen action (Zachmann et al., 1986). Again, eunuchoid skeleton, delayed bone age, and osteopenia could now be considered the effect of estrogen deprivation also in 17α-hydroxylase/17,20-lyase-deficient patients (Yanase et al., 1991). In conclusion the eunuchoid skeleton commonly seen in male hypogonadism is largely due to the lack of estrogen activity rather than of androgen activity (Faustini-Fustini et al., 1999).

6. Estrogen and bone mineral density

Along with skeletal maturation and progressive epiphyseal closure, the mineralization of the growing skeleton rapidly increases during puberty, especially in cancellous bone. Studies on congenital estrogen-deficient men clearly demonstrate that this phenomenon is under the control of estrogens also in the human male (Faustini-Fustini et al., 1999).

During adulthood, sex steroids are considered necessary in order to maintain bone mass in both sexes. The prevailing view among physicians probably still remains that androgens maintain bone mass in males as well as estrogens do in females. In fact, till now, the current concept was that androgens and estrogens are the major determinant of bone mass in men (Finkelstein et al., 1996; Wang et al., 1996) and in women (Speroff et al., 1996), respectively.

The new concept that estrogens may be a possible mediator of the androgen effects on male bone comes from studies on congenital estrogen deficiency, a condition in which a severe demineralization of the skeleton is constantly present. Androgens alone are probably not able to promote a normal skeletal mineralization since a severe osteoporosis was reported in all the three adult patients described so far (Smith et al., 1994; Carani et al., 1997; Bilezikian et al., 1998). The efficacy of estrogen treatment in normalizing the bone mineral

density (BMD) in adult men with aromatase deficiency has been well established (Carani et al., 1997; Bilezikian et al., 1998). In a recent study concerning the effects of different doses of transdermal estradiol on BMD in a man with aromatase deficiency a dose-dependent effect of estrogen replacement therapy on BMD has also been suggested (Rochira et al., 2000).

Apart from congenital estrogen deficiency, there is growing recognition that estrogens are one of the major determinants of BMD in the human male. In male to female transsexuals a positive effect of estrogen treatment on BMD has been shown (Reutrakul et al., 1998), even after orchidectomy (Van Kesteren et al., 1998). Beyond the models of aromatase deficiency and of transsexuals, large population studies documented that estrogens are probably implicated in the maintenance of male bone mineralization during adulthood and aging. Accordingly, a strict correlation between bioavailable estrogens and BMD has been documented in the elderly (Greendale et al., 1997) and in adult men (Ongphiphadhanakul et al., 1998; Khosla et al., 1998). Also a major risk of vertebral fracture has been established in men with low bioavailable circulating estrogens (Barret-Connor et al., 2000). In men with idiopathic osteoporosis and vertebral fractures, 6 months of testosterone treatment showed a positive correlation of BMD with increased levels of 17β estradiol, but not with serum testosterone (Anderson et al., 1997). Besides, in men with idiopathic osteoporosis low estrogen serum levels together with a decreased estradiol to sex hormone binding globulin ratio have been noted (Gillberg et al., 1999) and a reduction of ER-α expression in both osteoblasts and osteocytes has been documented (Braidman et al., 2000). Recently, also in Noonan's Syndrome estrogen deficiency has been suggested as the cause of osteopenia (Takagi et al., 2000).

The data suggest that estrogens explain at least part of the action on bone which were classically ascribed to androgens, even though a pure androgen action on bone is suggested by several authors (Orwoll, 1996; Hofbauer and Khosla, 1999). Thus, researchers are now in search of a better understanding about the direct role of androgens on bone. Particularly, it has been suggested that the sexual dimorphism of bone structure could be explained on the basis of different circulating androgen concentrations between the two sexes. In fact, in men bone mass is higher than in women and male bone is also stronger than that of females, probably as a consequence of differences in bone structure (bones are wider in men than women) (Bonjour et al., 1991). These gender differences in both cortical and cancellous bone mass are related to bone size in humans (Seeman, 1997; Loro et al., 2000). A possible role for testosterone in bone size establishment could be explained on the basis of differences in muscle mass between the two sexes, which account for a different mechanical action on bone and consequently for a sexual dimorphism of skeletal size resulting from adaptive growth to muscle mass. Besides, androgens are mainly involved in the stimulation of periosteal growth in rats, resulting in a greater amount of periosteal bone in male rats when compared with the female counterpart (Vanderschueren et al., 1992).

7. Estrogen replacement therapy in males with aromatase deficiency

Till now two different schedules of estrogen treatment have been used in aromatase-deficient men. Bilezikian et al. used increasing doses of conjugated estrogens (from 0.3 mg to 0.75 mg/day) during the first 12 months and a final dose of 0.75 mg/day for the subsequent 2 years was regarded as a substitutive dosage (Bilezikian et al., 1998). Carani et al. started with a high dose of transdermal estradiol and the dose was subsequently reduced in order to find the appropriate substitutive dosage also useful for a long-term treatment (Carani et al., 1997; Rochira et al., 2000).

It remains a matter of debate whether a high-dose of estradiol should be preferred to a lower one for starting estrogen replacement therapy in aromatase-deficient adult men.

In the treatment of constitutionally tall boys high doses of testosterone are recommended in order to rapidly obtain growth arrest as well as high doses of estrogens are commonly used in the treatment of constitutionally tall girls (Drop et al., 1998). Estrogens have a biphasic effect on growth plates: low doses of estradiol (4 μg daily) stimulate ulnar growth in boys, while higher doses (from 20 to 90 μg daily) probably lead to an inhibition of this process (Cutler Jr., 1997). High doses of estradiol should be therefore regarded as a good choice for starting estrogen replacement therapy in aromatase-deficient adult men. Accordingly, in a man with aromatase deficiency the treatment with 0.95 μg/kg weekly of transdermal estradiol for 6 months followed by 0.47 μg/kg weekly resulted in a complete epiphyseal closure within 9 months (Carani et al., 1997; Faustini-Fustini et al., 1999). By our experience, when the ephiphyseal closure is achieved, the dosage should be reduced in order to preserve a normal BMD and to maintain both serum estradiol and gonadotropins in the normal range (Rochira et al., 2000). Bilezikian et al. also obtained a complete epiphyseal closure in the other aromatase deficient adult man after 6 months of treatment, starting with a low-dose (0.3 mg daily) of conjugated estrogen which was progressively increased (0.75 mg daily) during the first 12 months of treatment (Bilezikian et al., 1998). The former would mime the physiologic changes during puberty in an adult man, while the latter is still conflicting with the serum estra-

diol levels above the normal range. Notwithstanding these differences in the schedule of treatment, epiphyseal closure was achieved shortly before in the patient described by Bilezikian et al.

It has been suggested that the minimal amount of exogenous estrogen necessary to maintain a normal BMD and hormones within the normal range may be 0.47 μg/kg weekly of transdermal estradiol without inducing undesirable side effects, such as gynecomastia and loss of libido (Rochira et al., 2000).

Recently, it has been demonstrated that, in women, the dose of estrogen needed to preserve bone mass and to decrease the risk of fractures may be lower than that commonly used in postmenopausal women (Genant et al., 1997; Cummings et al., 1998; Ettinger et al., 1998). Similarly, in an aromatase-deficient man a dose of estradiol lower than that commonly used in postmenopausal women resulted in bone health (Rochira et al., 2000). It could be speculated that the amount of estrogen needed to prevent osteoporosis is similar in both sexes and that, if a threshold for estradiol serum levels does exist below which BMD is impaired, this threshold probably does not differ between males and females.

Estrogen replacement therapy in men should be continued lifelong, because it will probably warrant a positive impact of estrogens on well being, cardiovascular diseases, and survival also in the human male (Sudhir and Komesaroff, 1999; Mendelsohn and Karas, 1999). In the future, this issue could be promising for male as well as for female health. Even though cases of aromatase deficiency in men will be rarely discovered, further studies on longer treatment of more patients are required before any correlation can be drawn regarding the optimal management of estrogen replacement therapy. Furthermore, the report of an aromatase-deficient male infant (Deladoëy et al., 1999) together with the possibility of making the diagnosis earlier on the basis of mother virilization during pregnancy raises the question of what age estrogen replacement therapy should be started in the aromatase-deficient male. At present, estrogen treatment of adult aromatase-deficient men should be started using high doses that should be lowered after epiphyseal closure is achieved. In order to prevent cardiovascular diseases and to improve well-being and survival, estrogen replacement treatment should be continued lifelong using the dosage that ensures a normal bone mineral density together with physiological LH, testosterone and estradiol serum levels in accordance with the patient's age.

8. Conclusions

In conclusion, estrogens seem to be the main sex steroid involved in the final phases of skeletal maturation and mineralization. This phenomenon takes longer in hypogonadal men and in patients with delayed puberty leading to eunuchoid body proportions and osteopenia as a result of insufficient availability of androgens for aromatization to estrogens.

Estrogen is the main sex steroids involved in the final phases of skeletal maturation (e.g. achievement and maintenance of peak bone mass, epiphyseal closure), even though the details of molecular estrogen action on epiphyseal closure and bone maturation remain to be elucidated. In fact, the exact mode and site of action of estrogens in the growing bone are not completely known. In the human, ERα has been localized in osteoblasts (Kusec et al., 1998) and both ERα and ERβ are expressed from human epiphyseal chondrocytes, suggesting that estrogens may have a direct osteogenic action (Kusec et al., 1998; Nilsson et al., 1999). There is also a general agreement that estrogens together with many other hormones (including GH, IGF-I, PTH, and PTH-related peptide), a great number of cytokines and growth factors (Ohlsson et al., 1998) are of crucial importance for postnatal longitudinal bone growth. It remains to be established in details the mechanism of estrogen action in the skeletal maturation process. We do not still know whether estrogens act directly on bone tissue or whether they modulate a more complex hormonal (endocrine and/or paracrine) network, including cytokines and growth factors. Recently a key role for the angiogenic vascular endothelial growth factor (VEGF) in the longitudinal bone growth and ossification process of the growth plate has been suggested (Gerber et al., 1999). A role for estrogens on direct VEGF modulation in bone tissue could not be excluded since estrogens modulate at least in part VEGF synthesis and secretion in several tissue (Agrawal et al., 2000). However, further studies are required in order to prove this fascinating hypothesis, endocrinology of growth plate remains a poorly understood issue.

At present, the relationship among estrogens, cytokines, growth factors and bone mineral density has been well characterized, improving our knowledge of the pathogenetic basis of osteoporosis. Estrogens promote osteoclastogenesis by increasing the synthesis of some cytokines and growth factors (mainly interleukin-6, interleukin-11, receptor for activator of nuclear factor-κβ [RANK]-ligand and the RANK-receptor) that regulates osteoclast differentiation (for reviews see Manolagas and Jilka, 2000; Rodan and Martin, 2000). As a result, the increase in the osteoclast number and activity account for elevated bone resorption and bone loss. Besides, this new field of research opens new perspectives in the treatment of osteoporosis based on the development of antagonists of cytokines receptors (e.g. RANK-receptor) (Rodan and Martin, 2000) and of selective estrogen receptor modulators as well (Ke et al., 2000).

In conclusion, several lines of evidence support the view that estrogens are required and mediate part of the actions of androgens on the bone at puberty and regulate bone homeostasis during adulthood in men.

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