GROWTH
Genetics & Hormones
Vol. 16 No. 1 March 2000

Growth and Estrogen

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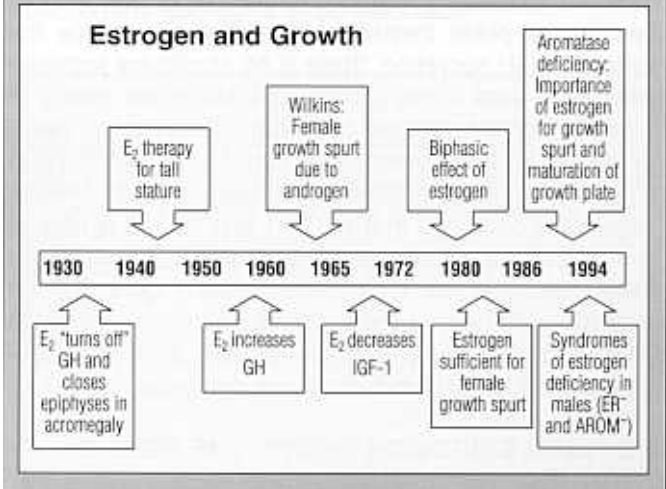
INTRODUCTION

During the past 5 years we have gained great insight into the critical role that estrogen plays in growth. This article reviews highlights of growth and estrogen research of the past 65 years (Figure 1), points out a number of earlier misconceptions, and culminates in the identification of "experiments of nature" that have revolutionized our understanding of the role that estrogen plays in linear growth.

Figure 1



Time line indicating the highlights in estrogen and growth research over the past 65 years. AROM⁻, aromatase deficient; E_2, estradiol; ER⁻, estrogen receptor deficient; GH, growth hormone; IGF-1, insulin-like growth factor 1.

GROWTH-INHIBITING EFFECTS OF ESTROGEN: THE EARLY YEARS

Children with precocious puberty have short stature as adults as a result of premature epiphyseal closure. Furthermore, in the absence of gonadal steroids, the epiphyses remain open and growth continues. The conclusion from these observations was that gonadal steroids were responsible for closing the epiphyses. Animal studies performed by Zondec in the 1930s revealed that estrogen had growth-inhibiting and growth hormone (GH) antagonistic properties.[1] As a result, it was concluded that estrogen, in addition to closing the epiphyses, "turned off" GH secretion.

As an extension of the assumption that gonadal steroids were responsible for turning off GH secretion, it was assumed that children would have more circulating GH than adults, the notable exception being patients with acromegaly. Beginning in the 1930s, in an attempt to inhibit growth and turn off GH secretion, patients with acromegaly were treated with gonadal extracts[2,3]; then, when pure steroid hormones became available in the 1940s, patients with acromegaly were treated with estrogens and androgens. The results with testosterone were disappointing, but estrogen proved to be highly effective, which was taken as proof that estrogen turned off GH secretion.[4]

In This Issue

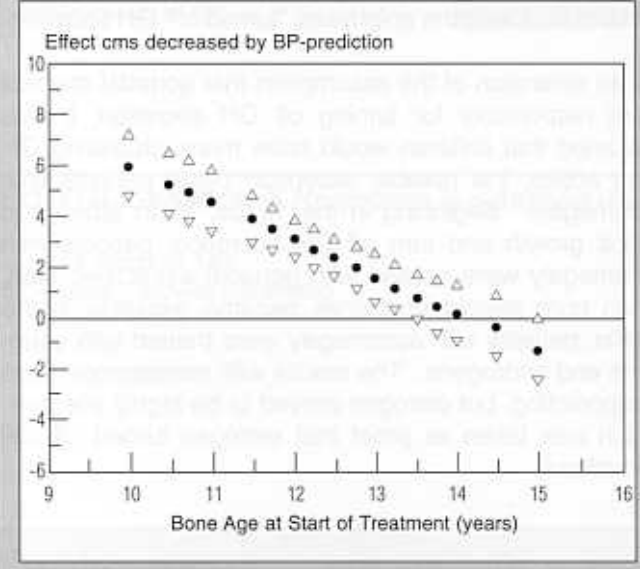
This educational activity is supported by an unrestricted educational grant from Genentech, Inc.

As an extension of the estrogen treatment of acromegalics, the late 1940s and early 1950s saw the start of estrogen treatment of excessive growth in adolescent girls.[5] A number of preparations have been used, including diethyl-stilbestrol, conjugated estrogens, injectable estrogen esters, and ethinyl estradiol. Ethinyl estradiol is the most widely used, and doses have decreased from 500 µg in the 1960s to 200 to 300 µg in the 1970s to 100 µg more recently.[6] Comparing studies is difficult because of the different preparations used, the different doses of estrogen administered, the varied duration of therapy, and the bone age at the start of treatment. Most studies show a growth-inhibiting effect that is inversely correlated with bone age at start of therapy (Figure 2).[6]

ESTROGEN DOES NOT "TURN OFF" GH SECRETION

It was only in the early 1960s that the radioimmunoassay was developed to measure physiologic levels of GH,[7,8] and it was a great surprise that young adults had higher levels of GH than children. Women also were noted to attain higher levels than men. Finally, in 1964 Rabkin and Frantz demonstrated that estrogen increases GH.[9] Therefore, the earlier assumption that gonadal steroids turned off GH secretion was incorrect.

Figure 2



Adjusted effect of estrogen therapy in adolescent females with tall stature. This representative study included 247 girls aged 12.7 ± 1.2 years. There were 88 controls and 159 treated subjects (90% were treated with ethinyl estradiol 200 µg). Duration of therapy was 1.9 ± 0.6 years. Mean length of follow-up was 10.9 years. Solid dots regression line; open triangles represent 95% confidence interval. Adjusted effect of estrogen (cm) = 20.22 – 1.44 × bone age (years).[30]

Reprinted with permission from de Waal WJ, et al.[30]

The immediate question that then arose was, "How, in an individual with open epiphyses, can estrogen slow growth while increasing GH?" The answer to this question came in 1972 from the elegant work of Wiedemann and Schwartz, who demonstrated that in acromegalics estrogen therapy (0.5 to 1.0 mg) caused a rapid fall in insulin-like growth factor 1 (IGF-1) but not GH. IGF-1 rose again when estrogen therapy was stopped.[10] In addition, they demonstrated that in patients with GH deficiency (GHD), estrogen therapy aborted the rise in IGF-1 that follows GH therapy.

Up until this point, only growth-inhibiting actions of estrogen had been demonstrated. In fact, in the Third Edition of Lawson Wilkin's textbook of endocrine disorders, published in 1965, the following statement appears: "Since estrogens have little or no effect upon nitrogen retention and in large doses may even inhibit it, ***it is probable that the adolescent growth spurt in females is due to adrenal androgens rather than to estrogen.***"[11]

GROWTH-PROMOTING EFFECTS OF ESTROGEN

Children with Turner syndrome lack estrogen and also lack a pubertal growth spurt. This observation prompted Ross, in 1983, to study the growth of patients with Turner syndrome in response to different doses of estrogen.[12] She studied 19 girls with Turner syndrome who received estradiol for 4 weeks in doses of 0, 50, 100, 200, 400, and 800 ng/kg/d in a double-blind manner. Patients received up to 3 monthly studies per year, and there was a 3-month washout period between monthly studies. She demonstrated a biphasic effect of estrogen on growth (Figure 3A). At low doses (100 ng/kg/d), there was a marked stimulatory effect on ulnar growth that disappeared at doses of 400 ng/kg/d and higher. This maximal stimulatory dose of estradiol (100 ng/kg/d) has been shown to increase the pulse amplitude of GH without affecting the pulse frequency.[13] However, despite the increase in GH secretion, there is no significant increase in the IGF-1 level at this growth-stimulating low dose.[12,13] Regarding longer duration of therapy, 5 µg ethinyl estradiol therapy daily in Turner syndrome (131 to 192 ng/kg/d) for up to 14 months resulted in increased growth velocity, again with no change in the IGF-1 levels.[14] It is only at the increased doses of estrogen, which have no effect on growth rate, that the IGF-1 levels rise (Figure 3B). It therefore appears that there also is a biphasic response of IGF-1 to estrogen in that intermediate levels of estrogen increase IGF-1 and high doses decrease IGF-1.[14]

THE FEMALE GROWTH SPURT: THE ROLE OF ESTROGEN

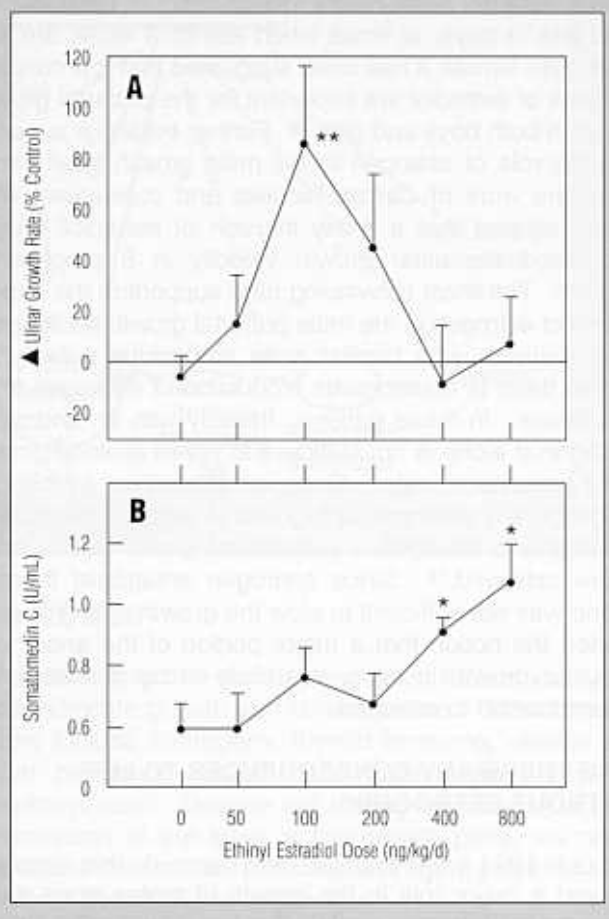
Given that estrogen can stimulate growth in females, it was important to question the assumption that the female growth spurt was due to androgen. Differentiating the roles that estrogen and androgen play in growth is very difficult

since estrogen and androgen are present in each sex. Testosterone is an obligatory intermediate in estradiol biosynthesis and aromatase, the enzyme that catalyzes the conversion of testosterone to estradiol, is found in males as well as females. In order to evaluate the role that estrogen plays in growth, without any influence from androgen, Zachmann (in 1986) studied 8 patients with androgen insensitivity.[15] These were individuals with disruption of their androgen receptors, and therefore they had only 1 functional sex steroid receptor. The pubertal peak height velocity occurred at a mean age of 12.7 years, closer to that of normal girls (12.4 years) than normal boys (13.9 years).[16] Mean peak height velocity was 7.4 cm/y, the same as in normal girls (7.3 cm/y) and lower than that in normal boys (9.3 cm/y). Therefore, estrogen alone, in the absence of androgens, is able to support a normal female pubertal growth spurt, both in magnitude and timing.

CAN ANDROGEN SUPPORT NORMAL PUBERTAL GROWTH IN FEMALES?

The answer to this question came in 1994 when Conte described a female patient with aromatase deficiency.[17] As a result of this deficiency, she had high levels of androgens and low levels of estrogens. At age 14 years, she had no breast development and no menarche. She had Tanner stage IV pubic hair, abundant axillary hair, acne, and an enlarged clitoris. Despite elevated androgens sufficient to produce virilization, she was short (height SDS of –1.5), had no growth spurt and, most remarkably, her bone age was delayed (10 years at chronologic age 14 years). With replacement therapy of 20 µg ethinyl estradiol, there was a striking decrease in her levels of androgens and gonadotropins, and she had a 13-cm pubertal growth spurt. Therefore, the assumption that the female pubertal growth spurt was due to androgens appeared to be incorrect.[11]

Figure 3



Relationship between dose of ethinyl estradiol and ulnar growth rate and serum somatomedin C.[12] *P <0.05; **P <0.025

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CME CERTIFICATION

The *GGH* Editorial Board is pleased to announce Category 1 credit for *GROWTH, Genetics, & Hormones* from the University of Virginia School of Medicine. This enduring material has been planned and produced in accordance with the ACCME Essentials.

Overview: This enduring material is designed to provide physicians and other health professionals with current research and clinical information essential to providing quality patient care to children with growth problems and genetic disorders.

Target Audience: This enduring material is designed for pediatricians, pediatric endocrinologists, pediatric geneticists, and family medicine physicians interested in pediatric growth, genetics, and endocrine issues.

Method of Physician Participation: Physicians can study each issue of *GROWTH, Genetics, & Hormones*, respond to the post-test self-evaluation questions, and request CME credit for each issue. The estimated length of time to complete this enduring material is 1 hour.

Learning Objectives: Through participation in this enduring materials series, the participant will have the opportunity to:

1. Apply current research and advances to the management of patient care for optimum clinical outcomes.
2. Utilize current research and clinical care issues to initiate discussions with colleagues with a focus toward increased awareness of current issues and controversies.
3. Conceptualize areas for future research in the field of growth and genetics.

THE ROLE OF ESTROGEN IN MALES

Since pubertal peak height velocity occurs early in girls and late in boys, at times when estradiol levels are low and quite similar, it has been suggested that low concentrations of estradiol are important for the pubertal growth spurt in both boys and girls.[18] Further evidence supporting the role of estrogen in the male growth spurt came from the work of Caruso-Nicoletti and colleagues, who demonstrated that a 4-day infusion of estradiol 4 μg/d increased the ulnar growth velocity in 5 prepubertal boys.[18] The most convincing data supporting the importance of estrogen in the male pubertal growth spurt came from patients with familial male precocious puberty, in whom there is autonomous production of androgen from the testes. In these patients, therapy with an androgen antagonist alone is not sufficient to revert skeletal growth to a prepubertal rate. Once an aromatase inhibitor is added to the androgen antagonist to block conversion of androgen to estrogen, a prepubertal growth rate is once more achieved.[19] Since androgen antagonist therapy alone was not sufficient to slow the growth rate, this supported the notion that a major portion of the androgen-induced growth in boys was likely to be mediated via aromatization to estrogen.

ARE THERE ANY CONSEQUENCES TO LIFE WITHOUT ESTROGEN?

Up until 1994 it was impossible to conclude that estrogen played a major role in the growth of males since there were no human male models that lacked estrogen action. However, in 1994, a man with complete estrogen resistance, caused by a disruptive mutation in the estrogen receptor gene,[20] was described. For the first time we were provided with the unique ability to evaluate the role played by androgen alone, in the absence of estrogen action. The man with estrogen resistance experienced normal prepubertal growth and normal onset of secondary sexual characteristics. He achieved his midparental target height (5 ft 10 inches) at age 16 years. Despite full masculinization, however, epiphyseal fusion had not occurred and, consequently, he continued to grow. At 28 years, he was 6 ft 8 inches with a bone age of 15 years, and he was growing at a growth velocity of approximately 1 cm/y.[20] One year later, a man with the identical phenotype was described.[21] The striking feature of this 24 year old also was tall stature and continued linear growth as a result of delayed skeletal maturation. He was 6 ft 8 inches with a bone age of 14 years at chronologic age 24 years. This man had a disruptive mutation in the aromatase gene and an inability to convert androgen to estrogen. Within 6 months of therapy with congugated estrogens (0.3 mg/d increased to 0.75 mg/d over the first year), linear growth ceased and his epiphyseal growth plates fused.[22] A second male with aromatase deficiency, with the same skeletal phenotype, also has been described (Table).[23]

Table
Syndromes of Estrogen Deficiency in Males

	Estrogen Resistance **Case**[20]	Aromatase Deficiency **Case 1**[21]	**Case 2**[23]
Age (y)	28	24	38
Height (cm)	204	204	190
Upper/lower ratio	0.88	0.84	0.85
Tall stature	yes	yes	yes
Continued linear growth	yes	yes	yes
Eunuchoid habitus	yes	yes	yes
Bone age	delayed	delayed	delayed
Bone mineral density	reduced	reduced	reduced

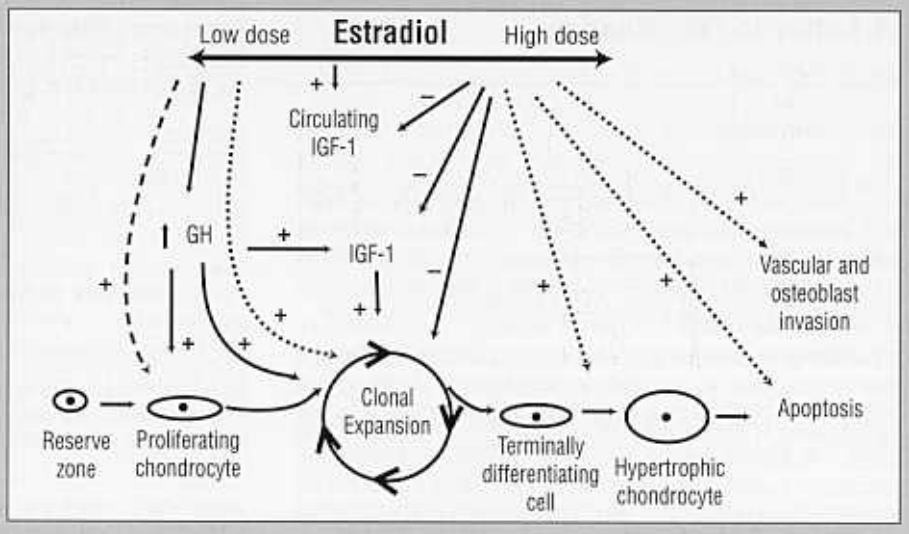
It is clear from the syndromes of estrogen deficiency that androgen, in the absence of estrogen, is relatively ineffectual in epiphyseal maturation.[24] However, it appears that androgen, in the environment of severe estrogen deficiency, is able to sustain linear growth despite arrested skeletal maturation. The estrogen-resistant and 2 aromatase-resistant males achieved their genetic potential for height at a normal age of 16 to 17 years, rather than at a later age, as would be expected with hypogonadal individuals. A possible explanation for the observed growth is that androgen, if not aromatized to estrogen, can stimulate growth directly at the level of the epiphyseal chondrocyte. In support of this are the observations made by Keenan and colleagues. They demonstrated that in short boys with delayed puberty, 5-dihydrotestosterone, a metabolite of testosterone and nonaromatizable androgen, induced and maintained an accelerated growth rate in spite of a 50% decline in integrated GH concentration and no change in IGF-1 level.[25]

All 3 males who lacked estrogen action have eunuchoid body proportions (Table), indicating relatively poor spinal growth (which is largely dependent on sex steroids) compared with limb growth. While it is tempting to speculate about the growth spurt of these individuals, there is insufficient longitudinal growth data on any of them to comment on the presence or absence of a growth spurt. Recently, a male infant with aromatase deficiency was described, and it will be highly informative to carefully follow his growth during his pubertal years.[26]

ESTROGEN ACTION AT THE GROWTH PLATE

As our clinical understanding increases, there is still much to learn about the mechanism of estrogen action at the growth plate. The growth plate is made up of chondrocytes, which are organized into layers. At the distal epiphyseal ends, the chondroblast progenitor cells occur singly or in small clusters to form

Figure 4



Proposed mechanism of action of estrogen at the level of the growth plate. The solid arrows represent data that have been demonstrated and the interrupted arrows represent other possible effects. Refer to text in article. GH, growth hormone; IGF-1, insulin-like growth factor 1.

the reserve zone. The next zone is the proliferative zone, in which the chondrocytes undergo clonal expansion and form discrete columns. The proliferative chondrocyte then undergoes terminal differentiation to form the hypertrophic chondrocyte. The hypertrophic chondrocytes secrete matrix, which undergoes mineralization. The hypertrophic chondrocytes then undergo apoptosis, and finally there is vascular and osteoblast invasion, which reduces the size of the growth plate.

Figure 4 represents some of our understanding of estrogen's role in growth. Low-dose estrogen increases GH secretion and stimulates growth. According to the dual effector theory of Green and associates,[27] GH primes the resting chondrocyte, preparing it for clonal expansion under the influence of IGF-1. However, GH receptors are not confined to the resting chondrocytes in the reserve zone, and GH and IGF-1 have been shown to exert their effect at each stage of differentiation.[28] The presence of estrogen receptor α in all populations of chondrocytes suggests that estrogen may have a direct role on the chondrocyte to stimulate growth.

High doses of estrogen inhibit growth by decreasing IGF-1 and inhibiting cell proliferation in the hypertrophic zone. The inhibition of clonal expansion by estrogen is not overcome by the addition of GH or IGF-1, suggesting that it is mediated directly by estrogen.[29] High-dose estrogen also may inhibit growth by inducing terminal differentiation of proliferating chondrocytes, apoptosis of hypertrophic chondrocytes, and vascular and osteoblast invasion into the growth plate.

CONCLUSION

Estrogen is only one of many important factors involved in chondrocyte growth and differentiation. Other critical factors include androgens, thyroid hormone, vitamin D, Indian hedgehog protein, and parathyroid hormone receptor protein. Despite our limited knowledge of the mechanisms at the level of the growth plate, we now appreciate the critical role that estrogen plays in the growth of both females and males.

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