Pituitary-Gonadal Relations in Female Children and Adolescents

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Extract

Data are provided which concern daytime levels of circulating follicle-stimulating hormone (FSH), luteinizing hormone (LH), and estradiol and progesterone in healthy female children and adolescents. These findings are correlated with sexual and skeletal maturation. Serum estradiol and testosterone levels were low in childhood, but rose to adult levels coincident with the appearance of secondary sexual characteristics. Serum LH levels did not rise above prepubertal levels until these characteristics were already apparent. Serum FSH levels showed a biphasic pattern with age; widely scattered values, some into the adult range, were seen in infancy. Mean FSH levels then declined throughout childhood until the onset of clinical puberty at which time they increased again to adult levels. Serum progesterone concentrations remained low (below 150 ng/100 ml) until menarche. Levels of serum progesterone which suggested possible ovulation and corpus luteum formation (over 200 ng/100 ml) were found in 9 of 30 girls who were less than 3 years postmenarchal.

Speculation

The finding of high serum FSH levels in female, but not in male, infants suggests that the infantile ovary may be relatively resistant to FSH stimulation. The decline in FSH levels during early childhood may represent increasing ovarian maturation, at least in terms of ability to secrete as yet unidentified gonadotropin-inhibiting feedback substance (or substances). The onset of female puberty appears to be heralded by increasing FSH secretion; although nocturnal LH release may be characteristic of early puberty, daytime elevation of this hormone is not seen until midpuberty. The surprising finding of luteal range serum progesterone levels in many early postmenarchal girls suggests that anovulatory cycles may not be characteristic of these years. The reported relative infertility of adolescent girls may relate to factors other than ovulation (such as a short corpus luteum life-span, defects in sperm capacitation, or impaired blastocyst implantation).

Introduction

In recent years the availability of sensitive and specific isotope displacement assays for gonadotropins and sex steroids has made possible more precise analysis of pi-

uitary-gonadal function during human development. We have recently reported data on levels of circulating FSH, LH, and testosterone in male children and adolescents [14]. It is the purpose of this report to provide similar data which concerns serum concentrations of

gonadotropins and gonadal steroids in healthy girls, and to correlate these findings with stages of sexual maturation.

Materials and Methods

Subjects

The 185 girls in the study included school children, ambulatory patients at the Children's Hospital, and student nurses. All were judged to be free of endocrine disease by history and physical examination. Informed consent for examination and venipuncture was obtained from each subject, her guardian, or both. Each girl's adolescent development was graded by secondary sex characteristics alone, in accordance with the following criteria: *stage P₁*, preadolescent; *stage P₂*, palpable subareolar breast tissue, sparse labial hair, or both; *stage P₃*, breast diameter 2–11 cm and hair which covered the pubes; *stage P₄*, breast tissue and pubic hair which approached the adult in amount and configuration, and axillary hair present. In this group, *stage P₄* breast development corresponds to *stage B₄* and *B₅* of Tanner [12]. A detailed menstrual history was obtained from all postmenarchal subjects. Venous blood was obtained between 9:00 and 11:00 AM on the examination day and the serum was stored at –20° until analysis. Skeletal maturation was determined from a wrist and hand radiograph in girls aged 6–18 using the standards of Tanner *et al.* [13].

Laboratory Methods

Measurements of FSH and LH were made in duplicate by double antibody radioimmunoassay [2, 3] in

sample volumes of 0.2 ml serum for adults and 0.4 ml for children. The human pituitary gonadotropin standard, LER-907 [20], was used in both assays. (The FSH and LH data in this report may be converted to milli-international units 2nd International Reference Preparation-human menopausal gonadotropin per milliliter by multiplication of the FSH value by 0.5 and the LH value by 4.5.) The minimum levels detectable in a 0.4-ml sample were 3.1 and 0.8 µg/100 ml for FSH and LH, respectively. The coefficients of variance for duplicate determinations were ±5% for FSH and ±6% for LH. Testosterone was measured in 2 ml serum by a competitive protein-binding technique [16] with a sensitivity of 8 ng/100 ml and a coefficient of intra-assay variance of ±7%. Estradiol was measured in 2 ml serum by a radioimmunoassay method [17] with a minimum detectable level of 0.5–1.0 ng/100 ml and a coefficient of intra-assay variance of ±7%. When sufficient sample was available, progesterone was also measured in 2 ml serum, using a competitive protein-binding assay [8] with a sensitivity of 5 ng/100 ml and a coefficient of intra-assay variation of ±8%. With this assay, ovulating women during the luteal phase of the menstrual cycle show serum progesterone concentrations in excess of 200 ng/100 ml, with peak values of approximately 2,500 ng/100 ml.

Results

The results are summarized in Table I by age and in Table II in accordance with the stage of sexual development.

Table I. Hormone levels in girls by age¹

Age, yr	n	Puberty stage ²	Bone maturation score ³	Serum				
				FSH, µg/100 ml ⁴	LH, µg/100 ml ⁴	Testosterone, ng/100 ml	Estradiol, ng/100 ml	Progesterone, ng/100 ml
0.5–2.0	17	P ₁	Not done	12.2 ± 2.3	2.1 ± 0.2	14 ± 2	All <0.5	Not done
2.1–4.0	6	P ₁	Not done	8.6 ± 1.9	1.6 ± 0.2	16 ± 2	All <0.5	Not done
4.1–6.0	11	P ₁	Not done	7.5 ± 1.2	2.2 ± 0.2	16 ± 2	All <0.5	Not done
6.1–8.0	25	P ₁	284 ± 12	6.1 ± 0.3	1.8 ± 0.1	21 ± 2	<0.5 (0–1.7)	<5 (<5–23)
8.1–10.0	25	P ₁	388 ± 20	7.0 ± 0.6	2.3 ± 0.2	24 ± 3	<0.5 (0–3.1)	6 (<5–52)
10.1–12.0	36	P ₂	575 ± 22	11.2 ± 0.7	2.5 ± 0.2	35 ± 3	1.6 ± 0.4	8 (<5–55)
12.1–14.0	21	P ₄	860 ± 28	13.8 ± 0.9	4.5 ± 0.5	37 ± 3	4.2 ± 1.2	37 (10–450)
14.1–16.0	17	P ₄	934 ± 13	18.9 ± 2.7	8.7 ± 2.4	52 ± 4	8.6 ± 1.5	289 (<5–1333)
16.1–18.0	6	P ₄	975 ± 11	22.2 ± 2.9	9.4 ± 1.9	51 ± 6	2.2 ± 0.7	161 (<5–952)
18.1–25.0	21	P ₄	Not done	17.6 ± 1.8	7.0 ± 1.0	57 ± 3	8.6 ± 1.2	Not done

¹ Data are reported either as the mean ± SE or as median (range). FSH: follicle-stimulating hormone; LH: luteinizing hormone

² Refers to the most common puberty stage in each age group.

³ By method of Tanner *et al.* [13].

⁴ LER-907 standard.

Table II. Hormone levels in girls by sex stage¹

Pu- berty stage	n	Num- ber post- men- archal	Age, yr	Bone maturation score	Serum				
					FSH, $\mu\text{g}/100\text{ ml}$	LH, $\mu\text{g}/100\text{ ml}$	Testosterone, $\text{ng}/100\text{ ml}$	Estradiol, $\text{ng}/100\text{ ml}$	Progesterone, $\text{ng}/100\text{ ml}$
P_1	82	0	7.0 (0.5-12.5)	<500	8.2 ± 0.6	2.1 ± 0.1	19 ± 1	<0.5 (<0.5-1.7)	<5 (<5-30)
P_2	23	0	10.7 (8.4-12.0)	498 ± 17	9.4 ± 0.9	2.1 ± 0.1	34 ± 3	1.3 (<0.5-6.6)	16 (<5-46)
P_3	28	7	11.8 (9.2-16.0)	721 ± 34	14.2 ± 1.1	3.7 ± 0.3	38 ± 2	2.9 (<0.5-10.5)	23 (<5-61)
P_4	52	50	16.0 (11.9-25.0)	933 ± 9	17.5 ± 1.2	7.6 ± 1.0	53 ± 2	7.6 ± 0.8	161 (<5-1333)

Data are reported either as mean $\pm$ SE or as median (range). FSH: follicle-stimulating hormone; LH: luteinizing hormone.

Fig. 1. Serum estradiol concentrations in girls at different ages. The shaded area shows the limit of sensitivity of the radioimmunoassay. ●: prepubertal girls; ▲: premenarchal girls with breast development; ○: postmenarchal girls. Values for subjects over age 16 are shown on the right as a function of days from the onset of the previous menses (LMP).

Preadolescent Girl (Stage P_1)

Preadolescent subjects ranged in age from 0.5-12.5 years; maximum bone age in these girls was 10.7 years. Estradiol was undetectable in the serum of most (Fig. 1), although levels of 1.0-1.7 ng/100 ml, which preceded the onset of breast development, were found in a few older stage P_1 subjects. Testosterone levels (Fig.

Fig. 2. Plasma testosterone concentrations in girls at different ages. The shaded area shows the limit of sensitivity of the assay. ●: prepubertal girls; ▲: premenarchal girls with pubic hair; ○: postmenarchal girls.

2) showed a similar pattern, as some values above 20 ng/100 ml were seen after age 7. Serum progesterone values in stage P_1 were all less than 30 ng/100 ml; no

progesterone was detected in most samples. Serum LH concentrations (Fig. 3) remained below $3.0 \mu\text{g}/100 \text{ ml}$ throughout childhood, with no apparent prepubertal rise. In contrast to the uniformly low LH levels seen in these girls, there was considerable scatter in serum FSH values, particularly during infancy when several values in the adult range were seen (Fig. 4). In later childhood, high values were not found and scatter was less marked; because of this, mean serum FSH levels declined during childhood ($r = -0.50$, $P = <0.001$).

Early Midadolescence (Stages P_2 and P_3)

The earliest physical signs of puberty were the appearance of labial hair (associated with a bone age of 9.1 years or more) or of a small bud of subareolar breast tissue (at a bone age of 9.5 years or more). A rise in mean serum levels of FSH, estradiol, and testoster-

Fig. 3. Serum luteinizing hormone concentrations in girls at different ages. The shaded area shows the limit of sensitivity of the assay. Symbols are as in Figure 1.

Fig. 4. Serum follicle-stimulating hormone concentrations in girls at different ages. The shaded area shows the limit of sensitivity of the assay. Symbols are as in Figure 1.

one was seen with the transition to stage P_2 . However, a rise in mean serum LH concentration above prepubertal levels was not seen until puberty stage P_3 , when secondary sexual characteristics were already well developed. Serum progesterone concentrations in the premenarchal adolescent girls of stages P_2 and P_3 were not significantly different from those seen in stage P_1 . Of the 28 stage P_3 girls, the 7 postmenarchal girls were the oldest and physically most mature (their mean bone age was 13.9 years, as compared with 12.3 years for stage P_3 as a whole). The gonadotropin, estradiol, and testosterone concentrations in these postmenarchal girls were in the normal adult range, but none had a serum progesterone level in the luteal phase range.

Menarche and Beyond (Stage P_4):

All but 2 of the 52 girls in stage P_4 were postmenarchal. All had hormone values in the normal adult female range and there was no evidence of a continuing increment in mean gonadotropin, estradiol, or tes-

Fig. 5. Serum progesterone concentrations in girls at different ages. Symbols are as in Figure 1. F: normal range in adult women during the follicular phase of the menstrual cycle; L: approximates the normal luteal phase range.

tosterone concentrations with age. The highest LH values were seen in samples taken 10–20 days after the last menses, reflecting the normal midcycle ovulatory rise of this hormone [9]. If all of the cycles in the postmenarchal girls were ovulatory, one would have expected to find a serum progesterone concentration greater than 200 ng/100 ml in just under one-half of the samples [9], inasmuch as these were drawn randomly in relation to the menstrual cycle. In fact, a serum progesterone value greater than 200 ng/100 ml was found in 9 of the 30 girls who were less than 3 years postmenarche (Fig. 5). Of four girls who were within 1 year postmenarche and in whom the preceding menses had occurred 14–28 days previously, two had serum progesterone concentrations in the luteal-phase range; the other two had apparently had an anovulatory cycle.

Discussion

Cross-sectional data cannot define the endocrine phenomena of adolescence for a particular girl, but can describe mean trends in hormone concentrations for the adolescent population. It is clear that for girls, as for boys [14], puberty is associated with rising serum

levels of pituitary gonadotropins. The earliest hormonal event of female puberty, as in the male, appears to be increased FSH secretion, with increased LH levels being delayed until secondary sexual development is moderately advanced. This is in general agreement with the cross-sectional data of Jenner *et al.* [6], Penny *et al.* [7], Sizonenko *et al.* [10], Yen and Vivic [18], and Yen *et al.* [19]. However, Boyar *et al.* [1] have recently described sleep-associated bursts of LH release as an early phenomenon of puberty, before waking levels of LH are increased; the sequence of these early hormonal events requires further study.

Two prominent differences from the gonadotropin pattern of male development are apparent: these are the wider range of FSH and LH values seen in mature females, and the considerable scatter in FSH levels found in some female infants [4]. The variability seen in the postmenarchal subjects reflects the appearance of menstrual rhythms in FSH and LH secretion. The scatter seen in FSH values in the infant females may also reflect rhythmic secretion at this time, inasmuch as a similar sex difference in FSH levels, with evidence for episodic secretion, has recently been demonstrated in infant chimpanzees [5]. One may speculate that bursts of FSH secretion may be characteristic of female infancy. The disappearance of such episodic FSH secretion in later childhood may be the result of increasing ovarian function at this time. Certainly, in the absence of a functioning gonad, marked hypersecretion of pituitary gonadotropins is a constant finding in childhood [15]. The identity of the ovarian secretion product responsible for such feedback inhibition of gonadotropin release is not known. It is clear from the present study that little estradiol, testosterone, or progesterone is being secreted by the prepubertal ovary.

As might be expected, the onset of breast development correlates well with rising concentrations of estradiol. The relatively wide scatter of estradiol values seen in the premenarchal adolescent girls (stages P_2 and P_3) may reflect episodic ovarian function before menarche. A concomitant rise in mean serum testosterone concentration was seen in association with pubarche. The origin of this circulating androgen (adrenal, ovarian, or both) and the stimulus for the observed rise cannot be determined from these data.

The serum progesterone data in this report suggest that regular ovulatory cycles may be established within a year after menarche. However the exact temporal relation of ovulation to menarche and the frequency of successful corpus luteum formation in the early postmenarchal years can only be established by more

detailed study. The possibility must be considered that the relative infertility of the adolescent may result from follicular luteinization without ovulation or from some other menstrual variation such as the "short luteal phase" [11].

Summary

Circulating daytime levels of FSH, LH, estradiol, testosterone, and progesterone were measured in 185 healthy females aged 0.5–25 years. These data were correlated with chronologic age, the stage of pubertal maturation, and bone maturation. The data demonstrate that clinical puberty is accompanied by rising mean levels of circulating FSH, estradiol, and testosterone, whereas rising LH levels were not seen until secondary sexual characteristics were well developed. Serum progesterone concentrations in the luteal (post-ovulatory) range were seen in 9 of 30 girls who were less than 3 years postmenarchal, which suggested that anovulatory menstrual cycles may be less frequent at this time than was thought previously. The unexpected finding of serum FSH concentrations during infancy which were higher than those in later childhood suggests that the infantile ovary may be relatively refractory to gonadotropin stimulation, at least in terms of its capacity to secrete materials with pituitary-feedback qualities.

References and Notes

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