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High-Frequency FSH and LH Pulses in Obese Menopausal Women

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We studied the pulsatile luteinizing hormone (LH) and follicle-stimulating hormone (FSH) secretory patterns, at early or intermediate years of menopause in seven normal women with different degrees of obesity, taking blood samples every minute for 40 min to 2 h. The hormones were assayed with an immunoradiometric assay (IRMA) system, analyzing with the cluster pulse algorithm. All women showed hormone pulses every 8–10 min. In five of them were found periods of discrete pulses with oscillations of high amplitude alternating with periods of pulses of low amplitude. In two cases, the high-frequency oscillatory pattern with low amplitude was found around low mean levels of 22.8 and 25.7 IU/L. The LH oscillatory pattern also had a high frequency, but at a lower level, giving a high FSH/LH ratio. The coincidence index of FSH with LH peaks was 76.6%. We concluded that at menopause, the frequency of FSH and LH secretion increases with a high FSH/LH ratio. Obese menopausal women may have the same high-frequency oscillatory patterns, but at low levels.

Key Words: FSH; LH; menopause; pulsatile secretion; obesity.

Introduction

Gonadotrophins are secreted in an intermittent fashion, in response to the hypothalamic pulsatile drive mediated by gonadotrophin-releasing hormone (GnRH) *(1,2)*. The pulses of GnRH are under the control of a pulse generator located at the hypothalamus *(3)*. During the menstrual cycle pulses of follicle-stimulating hormone (FSH) and luteinizing hormone (LH) are released at about 30-min intervals, with a high degree of coincidence *(4,5)*. At menopause, gonadotrophins are secreted unrestrained by the ovarian feedback, but the conditions of secretion are not similar for all menopausal women. FSH may increase with women's smoking habits, and decrease in obese menopausal women *(6)*. At menopause, the gonadotrophin pulsatile pattern secretion, increases in amplitude and frequency *(7)*, but the variations of the patterns among women are not well established. Different results on pulse frequency have been reported according to the frequency of sampling *(8,9)*. Considering that studies on cyclic profiles of hormone secretion and frequent sampling schemes may reveal different patterns of secretion *(2)*, we carried out this study to investigate the pulsatile patterns of LH and FSH secretion taking blood samples every minute in women at early or intermediate years of menopause. Considering the inverse relationship of FSH levels with obesity, we studied seven women with different values of body mass index (BMI).

Results

The characteristics of the patients are shown in Table 1, All had 10 or less years since menopause, and patients 4 and 5 had severe obesity (BMI, 37.7 and 42.9). Cases 4, 6, and 7 had vasomotor symptoms, as well as moderate symptoms of depression and anxiety. The hormone profile of the patients is shown in Figs. 1–7, and the data of pulse analysis are shown in Table 2. The analysis of peaks showed that the number of pulses, the mean interval between peaks, and mean peak width were similar for the seven studies.

In a global analysis of the general pulse pattern, it was surprising to find 5.2 (SD 1.0) peaks of FSH/h, with a mean interval between peaks of only 9.1 (SD 1.0) min. The general hormone secretory profile in cases 1, 2, 3, and 6 showed several clearly defined pulses with an amplitude (from close to 120 IU/L to decreases close to 30 IU/L) alternating with periods having pulses of decreased amplitude, but with similar duration. In contrast, cases 4 and 5, the women with larger BMI values, had lower mean FSH values (22.8 and 25.7 IU/L) than those expected for menopause and pulses with decreased amplitude (mean peak heights of 24.9 and 33.6). Case 7, also an obese woman (BMI, 32.2), showed an intermediate pattern with mean FSH values of 49.9, and mean peak height of 57.7.

The LH profile during the course of the study had peaks of lower amplitude in all cases as compared with those of FSH. The mean values were lower for cases 3 and 5, and the mean peak width was lower for case 4.

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Table 1
Characteristics of the Women in the Study

	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6	Case 7
Age	49	55	46	48	54	56	52
Age at menarche	13	13	12	11	13	14	14
Years since menopause	2	3	2	4	10	6	8
BMI	32.5	30.8	31.5	37.7	42.9	33.1	32.3
Abdomen/hip ratio	0.96	1.02	1.04	1.14	1.42	0.98	1.25
Waist/hip ratio	0.86	0.88	0.96	1.00	0.78	0.80	0.82
Parity	2	9	1	1	9	11	6

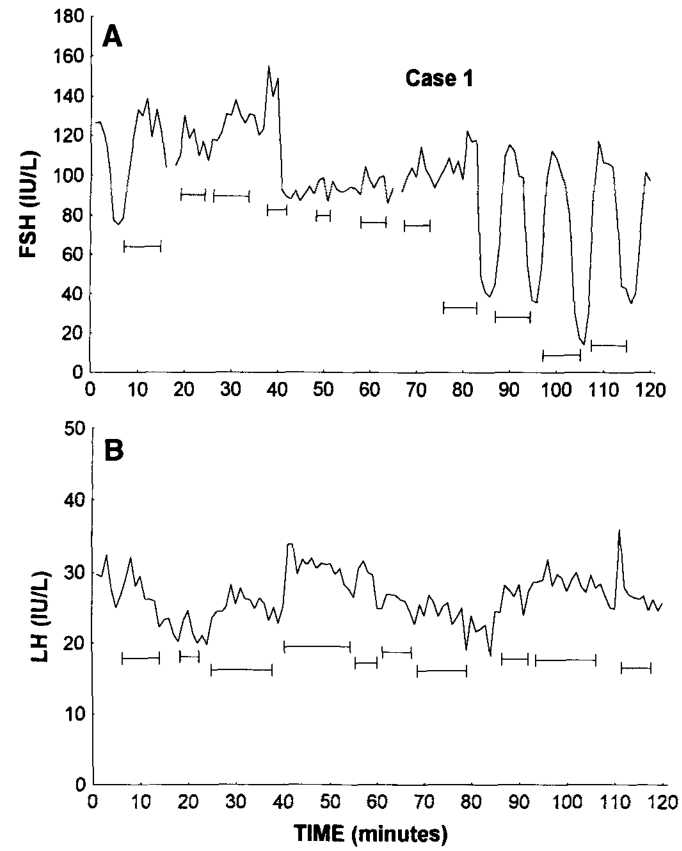

Fig. 1. FSH (**A**) and LH (**B**) secretory patterns of case 1. Horizontal bars indicate the wide peaks.

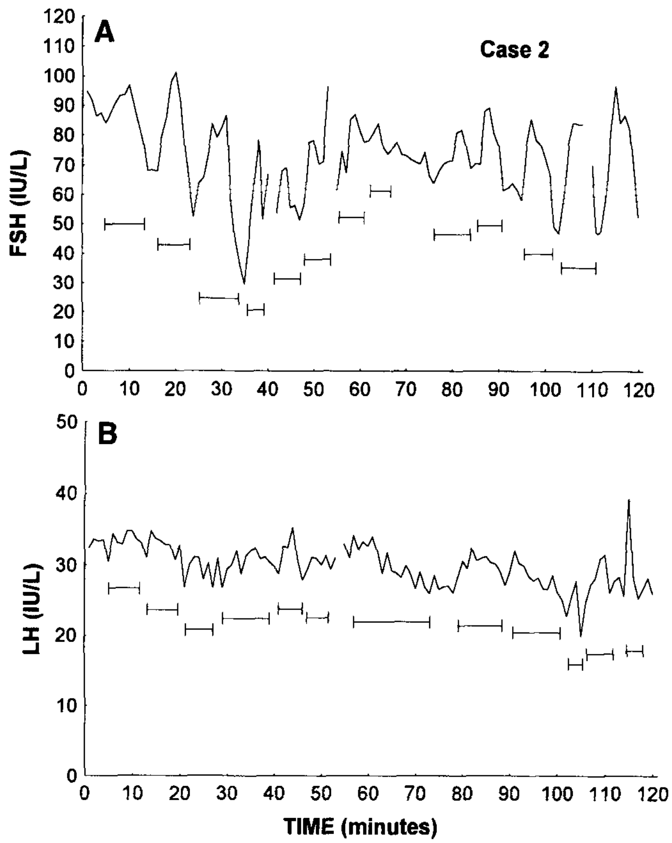

Fig. 2. FSH (**A**) and LH (**B**) secretory patterns of case 2. Horizontal bars indicate the wide peaks.

The coincidence rate of FSH with LH peaks was 8/11 for case one, 9/12 for case 2, 8/11 for case 3, and 11/13 for case 4. For the four cases studied during 120 min was 36/47 = 76.6%.

Discussion

During menopause, with the exhaustion of ovarian follicles, the ovarian capacity to produce estrogens and inhibin, the main feedback products for gonadotrophin regulation, is impaired. As a consequence, the gonadotrophin secretion increases unrestrained by ovarian regulation in an open-loop mechanism. Nevertheless, the possible influences of other central or peripheral factors have not been properly defined. The patterns of gonadotrophin secretion have been studied by several investigators *(7,10)*. Some of them have proposed that at menopause decreases the frequency of FSH *(11)*, but Reame et al. *(12)* report an increased pulse frequency in premenopausal women. The work of Kletzky et al. *(8)* reports that FSH pulses followed a single pattern with a pulse frequency of 1.7/h, whereas LH showed two different patterns, with an average of 1.1 pulses/h, sampling every 3 min. Rossmanith et al. *(9)* found 4.4 pulses of LH and 2.0 pulses of FSH in 8 h, in elder postmenopausal women using a sampling scheme every 10 min. Iranmanesh et al. *(13)* have proposed that intensive sampling procedure increases the probability for detection of high-frequency pulses.

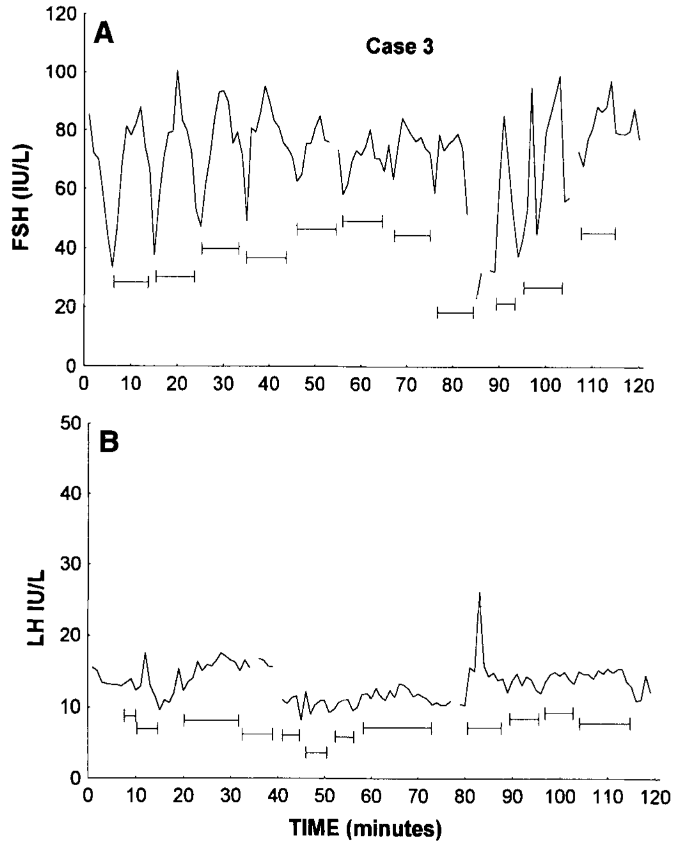

Fig. 3. FSH (**A**) and LH (**B**) secretory patterns of case 3. Horizontal bars indicate the wide peaks.

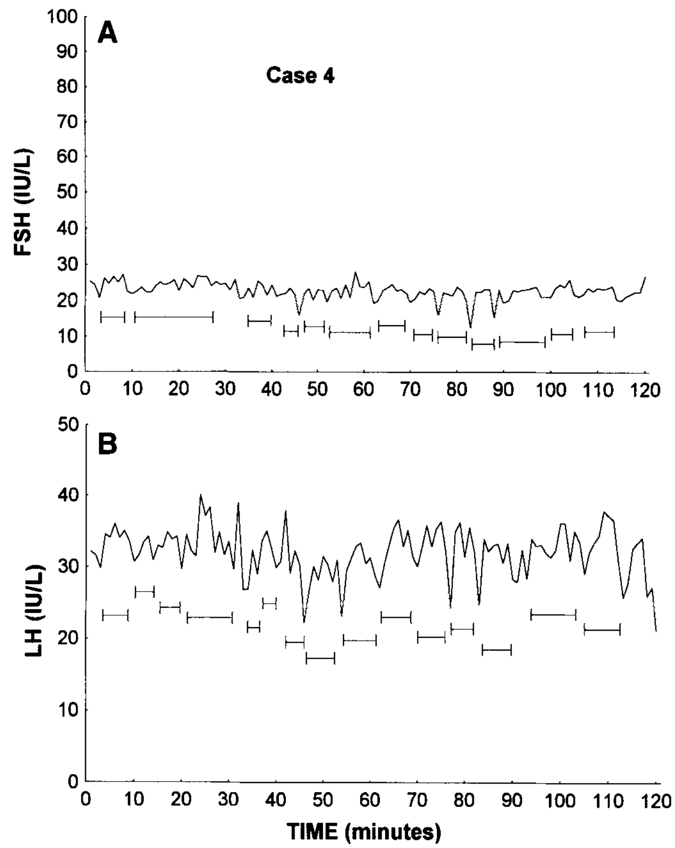

Fig. 4. FSH (**A**) and LH (**B**) secretory patterns of case 4. Horizontal bars indicate the wide peaks.

In this work, we studied postmenopausal women, taking samples every minute for 2 h and found an FSH pulsatile pattern of high frequency, with pulses every 9 min. Several patients showed a surprising sharp decline in gonadotrophin blood levels. The high reproducibility of the assay, as well as the remarkable congruence with neighbor points, makes it unlikely that they results from assay inaccuracies. The possibility that they are artifacts from dilution during the sampling procedure was ruled out, because it was a direct blood sample without the use of fluids for maintenance of permeability of the venous catheter.

The FSH secretory dynamics has been confounded by its heterogenicity *(14)*. Various works have reported that FSH isoforms secreted in pulses have a much shorter half-life compared with that of FSH secreted between pulses *(15,16)*. The nature of this short half-life secretory material has not been elucidated. The high rates of coincidence in the pulses of LH and FSH, are in agreement with the concept that the increased frequency in gonadotrophin secretions are the consequence of a pulsatile hypothalamic drive of GnRH secretion. These results are at variance with those found with less frequent sampling procedures *(8)*. It was surprising to find such short-lived pulses of FSH considering the long metabolic clearance rate of FSH. However, endogenous immunoreactive FSH has a greatly increased clearance rate as compared with exogenously administered FSH *(14)*.

In our study, except for case 4, FSH values were higher than those of LH. This may be explained because feedback ovarian signals are given by steroid hormones and inhibin, and the latter, a product of the ovarian follicle, is essentially FSH-specific. Reame et al. *(12)* studied women over 40 yr of age, and they found sustained elevation of the FSH/LH ratio at premenopausal years, with increased frequency and amplitude of LH pulses.

It is of interest to note that in four of the patients, we found periods with a high-amplitude pulsatile pattern, alternating with other intervals with pulses showing similar frequency but low amplitude. This may indicate that the pulsatile hypothalamic drive may interact with other peripheral influences not yet completely characterized, such as steroids with biological action, derived from the peripheral conversion of adrenal steroids. It is also possible that suprahypothalamic influences may also distort the episodic pattern given by the pulse generator, which after released from the ovarian feedback influences, may become more liable for influences, such as stress stimuli.

In cases 4 and 5, the menopausal women with the higher BMI, the pulsatile pattern of FSH showed a similar high frequency, but with low amplitude around low mean levels, as compared with the expected range for menopausal women. This is in agreement with our previous finding that FSH values are lower in obese menopausal women *(6)*. We

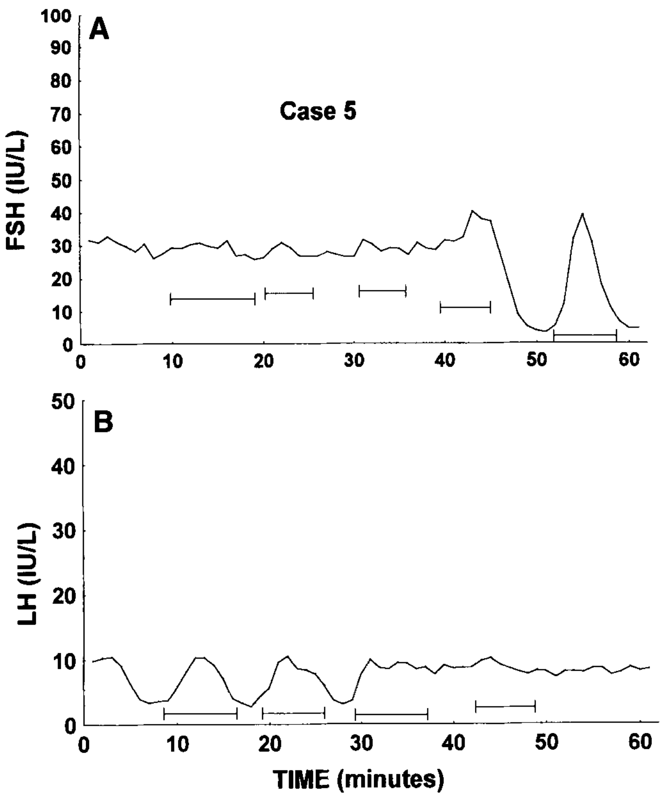

Fig. 5. FSH (**A**) and LH (**B**) secretory patterns of case 5. Horizontal bars indicate the wide peaks.

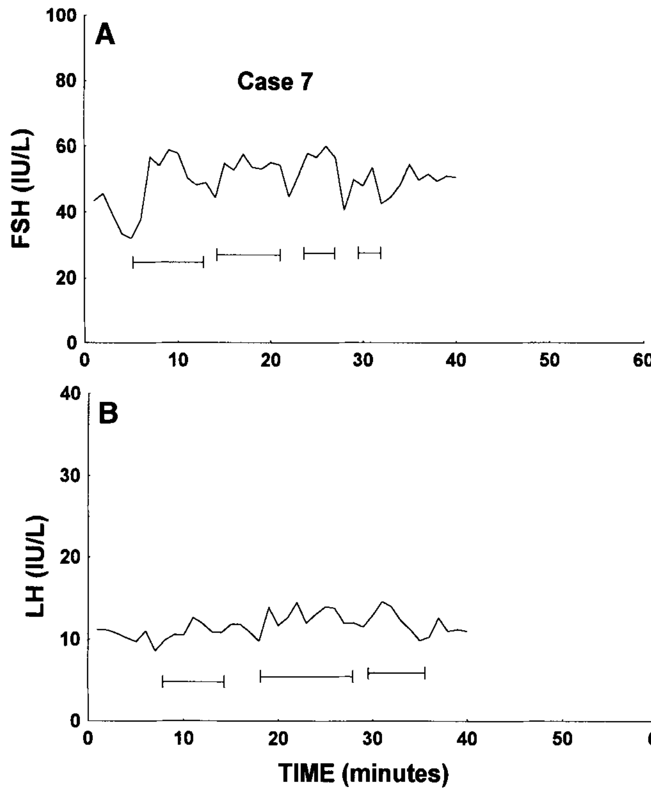

Fig. 7. FSH (**A**) and LH (**B**) secretory patterns of case 7. Horizontal bars indicate the wide peaks.

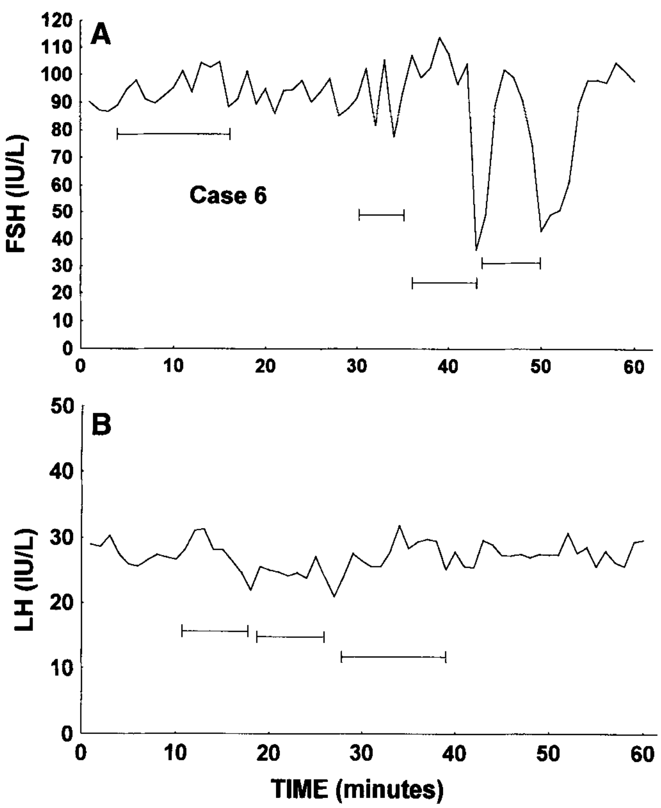

Fig. 6. FSH (**A**) and LH (**B**) secretory patterns of case 6, with sampling every minute. Horizontal bars indicate the wide peaks detected by the Cluster Detection Algorithm.

interpreted this event as the result of the peripheral overproduction of estrogens and androgens *(17,18)*. Furthermore, O'Dea et al. *(20)* found that dietary weight loss induces an increase in gonadotrophin secretion in obese postmenopausal women. This phenomenon underlines the importance of a careful evaluation of FSH secretion in obese menopausal women for the assessment of their risk factors and for the decision to prescribe hormone replacement therapy. Obese women have higher risk for endometrial cancer *(20, 21)*, but lower risk for osteoporosis *(22)*. Both events are possibly related to a higher level of estrogen exposure. We concluded that the early or intermediate years of menopause increase the frequency of FSH and LH pulses with a high FSH/LH ratio, and obese menopausal women may have high-frequency oscillatory patterns at low levels.

Materials and Methods

Subjects

We studied seven women at early or intermediate postmenopausal years, according to the following criteria for inclusion: Women with 1–10 yr of menopause, nonsmokers, nonhysterectomized, with a history of regular menses, without hormone replacement therapy for at least 6 mo before study, without apparent infectious, metabolic, or tumoral disease. The volunteers, not seeking treatment, were recruited by means of invitation in public areas of the city.

Table 2
Results for the Cluster Analysis of LH and FSH Pulses (Mean ± SD)

	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6	Case 7
FSH							
Number of peaks/hour	5.5	6	5.5	6.5	4	4	4
Mean values	97.5 ± 28.7	73.2 ± 13.9	71.7 ± 15.7	22.8 ± 2.4	25.7 ± 9.3	90.4 ± 16.2	49.9 ± 6.9
Mean interval between peaks (min)	9.7 ± 1.6	8.7 ± 3.6	10.2 ± 1.7	8.7 ± 3.3	9.0 ± 3.0	10.3 ± 6.6	7.3 ± 2.08
Mean peak width (min)	7.0 ± 1.9	6.0 ± 1.6	7.6 ± 1.6	6.2 ± 3.9	6.5 ± 3.5	7.2 ± 3.6	5.0 ± 2.4
Peak height (IU/L)	122.6 ± 16.5	86.8 ± 8.7	89.8 ± 7.6	24.9 ± 1.6	33.6 ± 4.5	106.4 ± 5.1	57.5 ± 2.8
Peak height as % increase	242 ± 181	162 ± 46	203 ± 126	123 ± 8	124 ± 10.9	151 ± 58.7	137 ± 29.1
Area	360 ± 198	145 ± 80	214 ± 111	20 ± 10	65 ± 99	187 ± 174	56 ± 54
LH							
Number of peaks/hour	5	6	6	7.5	4	3	3
Mean values	26.6 ± 3.3	30.0 ± 2.9	13.2 ± 2.4	32.0 ± 3.4	7.6 ± 2.2	27.0 ± 2.2	11.6 ± 1.4
Mean interval between peaks (min)	11.4 ± 4.6	9.6 ± 5.8	9.2 ± 4.4	7.4 ± 3.1	10.7 ± 2.1	10.5 ± 2.1	10.0 ± 1.4
Mean peak width (min)	8.2 ± 4.3	7.0 ± 4.4	9.6 ± 5.0	5.3 ± 2.3	7.5 ± 1.7	8.3 ± 3.2	6.3 ± 2.3
Peak height (IU/L)	30 ± 4	33 ± 3	15 ± 5	36 ± 2	10.3 ± 0.2	30.1 ± 2.6	13.9 ± 1.1
Peak height as % increase	110 ± 4	133 ± 56	156 ± 114	121 ± 6	265 ± 102	125 ± 15	133 ± 8
Area	32 ± 27	54 ± 96	30 ± 36	23 ± 12	28 ± 17	36 ± 23	15 ± 7

The purpose of the study was explained to them, and they signed informed consent. The protocol was approved by the institutional ethical committee.

Data Collection

The following data were collected by means of a questionnaire: date of birth, age at menarche, parity, menstrual history, date of the last menses, use of medication, alcohol intake, smoking habits, drug abuse, and physical exercise, as well as symptoms and previous diagnosis. The weight and standing height were measured with indoor clothing, but without shoes. The trunk circumference ratios were measured, and the following somatometric indices were calculated: BMI (weight/height^2, Kg/m^2), waist/hip, and abdomen/hip ratios. Sitting blood pressure was taken after 10 min of rest, and every 20 min during the blood sampling.

Blood Sampling

After a 12-h fast, a heparinized indwelling accessory for venous infusion needle no. 19G was placed in the antecubital vein for serial sampling, and a 1-mL blood sample was drawn at 1-min intervals. The first sample was discarded, and subsequent samples were obtained without the use of fluids for the maintenance of permeability. The study was carried out for 2 h in the first four patients, for 60 min in the next two, and for 40 min in the seventh patient. Serum was obtained by centrifugation after clotting at room temperature, and stored at –20°C until hormone assays.

Hormone Assays

LH and FSH were measured by immunoradiometric assay (IRMA) (ICN Biomedicals, Costa Mesa, CA). For each series of samples, a pool of serum was obtained from three different regions of the assay, in order to calculate the intra-assay coefficient of variation. Coefficient of variation ranged from 5.4–7.4 for LH, and from 6.2–8.3 for FSH.

Pulse Analysis

For analysis, we used the Cluster pulse algorithm developed by Veldhuis and Johnson *(23)*. Clusters of two points were required for a pulse detection. Also, a nadir cluster size of 2 points was required for the detection of a decrease. The criteria for false-positive pulse detection was set at < 2.5%. The coincidence rate of FSH and LH peaks was considered when the maximum increase of peaks was separated by 4 min or less.

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