Impact of fatness, insulin, and gynecological age on luteinizing hormone secretory dynamics in adolescent females

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Objective: To study the link between fatness and gonadotropin secretion. Overweight status is linked to polycystic ovary syndrome (PCOS) in adolescents. We postulated that heavier adolescents without symptoms would secrete LH with: [1] increased pulse frequency (LHPF) and [2] exaggerated integrated concentrations (LHAUC).
Design: Cross-sectional.
Setting: General clinical research center.
Patient(s): Eighty-seven postmenarcheal cyclic adolescents from lean to overweight recruited during the follicular phase.
Intervention(s): Luteinizing hormone sampling: [1] every 10 minutes/24 hours; [2] at 20-minute intervals after a GnRH challenge.
Main Outcome Measure(s): The LHPF and LHAUC (calculated by the CLUSTER algorithm). Hormonal and metabolic covariates included percent body fat (PercentBF), insulin-like growth factor-I (IGF-I), fasting insulin, and the insulin resistance index HOMA-IR. The SAS software was used for analyses.
Result(s): The PercentBF and younger gynecological age predicted faster LHPF. Fatness was negatively linked to LHAUC, which was best predicted by PercentBF and IGF-1 in multivariate modeling ($R^2 = 0.25$). The PercentBF and insulin predicted a lower 20-minute LH response to GnRH.
Conclusion(s): [1] Higher adiposity and younger gynecological age predict rapid LHPF. [2] The early years after menarche represent a vulnerable window for an exaggerated LHPF with weight gain. [3] In healthy adolescents, higher adiposity is linked to lower LHAUC, thereby preserving pituitary stores. (Fertil Steril® 2010;94:221–9. ©2010 by American Society for Reproductive Medicine.)

Key Words: Fatness, insulin, testosterone, LH, FSH, GnRH stimulation, adolescents

Fatness accelerates the onset and the progression of puberty in adolescent women (1–4) but the relationships between adiposity and LH secretion characteristics during the transition years of adolescence to adulthood are not known. Nutritional status as a determinant of fertility has been reported since centuries ago, as heavier adolescent women were noted to be more fertile than their leaner counterparts (5, 6). In recent days, however, the magnitude of the obesity epidemic has brought to the forefront the paradox of the overweight, yet infertile young woman (7–9).

Obesity can alter fertility potential but this outcome is rarely sought-out during the adolescent years. In addition, fertility cannot be reliably assessed in adolescents because adolescence is a transitional time, one of instability in the hypothalamic-pituitary-ovary axis when fertility is being established and by definition cannot be assessed directly. Nonetheless, the onset of fertility disorders related to obesity begins during this transition. Because the adolescent population has grown heavier, the ranks of women affected with polycystic ovary syndrome (PCOS) have been swelling with seemingly younger patients presenting with the clinical triad of oligomenorrhea, insulin resistance, and excessive androgen concentrations (10, 11). The cause of PCOS, a common cause of infertility (12), remains unknown. Although sharp clinical contrasts can be drawn between PCOS-affected adolescents and their healthy counterparts (13), it has been difficult to delineate the natural history of this syndrome as it emerges in young women for lack of hormonal data referring to the years following menarche. Adolescents with PCOS are hypergonadotropic with LH levels increased two- to threefold above average follicular phase levels and following a secretion pattern of high pulse frequency (14). The relative increase of LH is often expressed relative to the other gonadotropin, FSH with a LH-to-FSH ratio greater than one (15). One question that remains unanswered is to what extent the clinical and metabolic characteristics that define PCOS modulate fertility in healthy, regularly cycling young women with no clinical evidence of PCOS. We thus sought to determine

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whether obesity would relate to LH secretion characteristics in healthy regularly cycling adolescent women across the weight spectrum from lean to overweight.

We have shown that body mass index (BMI), the most widely used clinical measure of obesity is of limited value in the assessment of metabolic risks in adolescent females, whereas body fat percentage is a better index of energy balance and sensitivity to insulin (16). Bearing in mind that in PCOS, LH secretion is typified by rapid pulse frequency (17–19) and markedly increased LH concentrations (14), we hypothesized that both LH pulse frequency and 24-hour integrated concentrations would increase with adiposity in healthy postpubertal females. As a reflection of fertility potential, we studied gonadotropin secretion in two ways: first, by sampling LH every 10 minutes over a 24-hour period, and second, by measuring the peak LH response to a GnRH challenge.

MATERIALS AND METHODS

The protocol was approved by the Institutional Review Board (IRB) of the University of Michigan Health System. Eighty-seven adolescent females 14–20 years with BMI from lean to overweight for age (based on growth charts issued by the National Institute for Health Statistics) (20) were studied a minimum of 2 years after the age at menarche as part of a study on fitness and weight control. The participants were recruited between 2000 and 2008 from local university campuses and high schools and were enrolled after signing an IRB-approved consent form. The maximal age of recruitment was 20 years; however, four participants reached their twenty first birthday before completing the study. Gynecological age was calculated as the number of years between the reported age at menarche and the time of the study. All participants were healthy, nonsmoking, and taking no medications. At the time of enrollment, an intake questionnaire was administered to screen for symptoms associated with conditions causing menstrual cycle disruption, such as hypothyroidism and hyperprolactinemia. Participants who reported cold intolerance, nipple discharge, constipation and depression, rapid weight gain or weight loss were eliminated from consideration. Other criteria of exclusion were any hormonal method of birth control for the past year, history of depression, and facial hair. After this initial screen, a dietary history was collected to screen for restrictive or aberrant eating behavior and confirm a history of appropriate caloric intake for age and size. For this purpose, a 3-day food diary was mailed in advance of the admission to the General Clinical Research Center. The dietary data collected prospectively were then complemented by a one-on-one interview with the General Clinical Research Center nutritionist. Nutrient calculations were performed using the Nutrition Data System for Research software, version 4.03, developed by the Nutrition Coordinating Center, University of Minnesota, Minneapolis, MN, Food and Nutrient Database 31, released November 2000. The average caloric intake was 1,837 $\pm$ 475 kcal/day, appropriate for sedentary to moderately active adolescents (21).

Experimental Design

Subjects and sampling period At the time of the admission, the physical examination was performed by a pediatric endocrinologist to further confirm the absence of hirsutism, defined as a male pattern of hair distribution (22) or associated clinical evidence of androgen excess such as cystic acne and temporal balding. All participants were asked about the regularity of their menstrual cycle as it would be assessed in a routine clinic visit. Eligible participants reported "cycling every calendar month" around the same day of the month. All the participants were interviewed twice, first by questionnaire at the time of enrollment, then again at the time of the admission to the General Clinical Research Center and all were assessed to have at least 10 cycles per year. In addition, because of the imprecision inherent to the reporting of menstrual cycle details in adolescents, blood sampling was postponed from all participants until the onset of a subsequent cycle. The adolescent women were instructed to contact the research team (by e-mail or by telephone) on the first day of the menstrual cycle immediately after their enrollment in the study. This allowed for scheduling of the admission during the follicular phase, on the weekend immediately after the onset of the menstrual cycle. The day of the cycle was recorded and was subsequently used as a variable to adjust for differences resulting from different days of data collection within the follicular phase. Follicular phase status was further confirmed by serum P measured on the night preceding the study and before proceeding with sample collection. By 8 AM, an intravenous catheter was inserted in the forearm and blood was sampled every 10 minutes. Follicle-stimulating hormone was measured every hour and LH was measured every 10 minutes to account for its rapid pulsatility. Total T and E_2 were measured in a single sample at the time of admission. At the end of the 24-hour sampling, 250 ng/kg of synthetic GnRH (Factrel; Ayerst Laboratories, Rouses Point, NY) was administered IV (23) and LH and FSH responses were measured at baseline, then 20, 40, and 60 minutes later. At midpoint during the study, GnRH became unavailable for purchase and thus stimulated gonadotropin data are only available in a subset of 48 participants.

Metabolic measures All participants were weighted on a digital scale and height was measured using a wall-mounted stadiometer. Body mass index was calculated as the weight in kilograms divided by height in meter squared. Relative body fat was assessed by dual roentgenogram absorptiometry using a total body scanner (model DPX-L; Lunar Radiation Corp., Madison, WI) (24). In addition to quantitating body fat or fatness, we used the adipocyte-derived leptin, which was measured hourly for 24 hours, as a biomarker of adiposity (25, 26). To document sensitivity to insulin, fasting insulin, glucose, and insulin-like growth factor-I (IGF-I) were measured after a supervised 8-hour overnight fast on the second morning of the 24-hour sampling period.