Reprinted from AMERICAN JOURNAL OF OBSTETRICS AND GYNECOLOGY, St. Louis
Vol. 164, No. 4, pp. 989-996, April, 1991, (Printed in the U.S.A.)

The induction of premature luteolysis in normal women—follicular phase luteinizing hormone secretion and corpus luteum function in the subsequent cycle

Michael R. Soules, MD,[a] William J. Bremner, MD, PhD,[b, c] Kristine D. Dahl, PhD,[b, c] Jean E. Rivier, PhD,[d] Wylie W. Vale, PhD,[d] and Donald K. Clifton, PhD[a]
Seattle, Washington, and La Jolla, California

Women with luteal phase deficiency have been shown to have an increased frequency of luteinizing hormone pulses in the early follicular phase of the menstrual cycle. Because progesterone is known to modulate luteinizing hormone secretion, it has been hypothesized that the decreased progesterone secretion in a previous luteal phase deficiency cycle could lead to the abnormal luteinizing hormone secretory pattern in the ensuing early follicular phase. With the possibility that the higher luteinizing hormone pulse frequency might lead to another deficient luteal phase, it becomes conceivable that luteal phase deficiency could be self-perpetuating. To test this hypothesis, luteal phase deficiency was induced in six normal women by decreasing luteinizing hormone support of the corpus luteum with a gonadotropin-releasing hormone antagonist Nal-Glu, administered twice daily beginning in the midluteal phase after a control cycle. During the antagonist-treated luteal phase, each subject met the predetermined criteria for induced luteal phase deficiency: a 33% or greater decrease in integrated progesterone from the control cycle and an integrated progesterone level <100 ng/ml per day. Luteinizing hormone secretion patterns were determined by frequent blood sampling performed every 10 minutes for 12 hours in the early follicular phase of the control cycle and the cycle after antagonist administration. Daily luteal progesterone levels were measured in the control, treatment, and posttreatment cycles. Each volunteer served as her own control. Standard parameters were compared between the control and posttreatment pulse studies in the early follicular phase: (1) luteinizing hormone pulse frequency was 9.5 ± 1.0 vs 10.0 ± 0.9 pulses/12 hours, control vs posttreatment, respectively, $p = 0.5$; (2) luteinizing hormone pulse amplitude was 11.0 ± 1.3 vs 12.0 ± 2.2 ng/ml, $p = 0.6$; and (3) luteinizing hormone mean level was 19.4 ± 2.3 vs 22.2 ± 3.3 ng/ml, $p = 0.1$. Corpus luteum function was also compared between the control and posttreatment cycles. Luteal phase length was 13.7 ± 0.6 vs 12.7 ± 0.6 days, $p = 0.08$. Integrated progesterone values were 136.9 ± 12.9 vs 130.5 ± 11.3 ng/ml per day, $p = 0.5$. Therefore no discernible abnormalities in early follicular luteinizing hormone secretions or corpus luteum secretion of progesterone occurred after an induced luteal phase deficiency cycle. We conclude that luteal phase deficiency is not self-perpetuating and that the rapid luteinizing hormone secretion pattern found in the early follicular phase in women with luteal phase deficiency is not a result of decreased progesterone levels in the preceding luteal phase, but rather appears to be a result of dysfunctional suprahypothalamic neuroendocrine modulation of the gonadotropin-releasing hormone pulse generator. (AM J OBSTET GYNECOL 1991;164:989-96.)

Key words: Luteal phase deficiency, progesterone, luteolysis, gonadotropin-releasing hormone antagonist, gonadotropin secretion, corpus luteum

From the Departments of Obstetrics and Gynecology[a] and Medicine,[b] University of Washington, Veterans Administration Medical Center,[c] and the Peptide Biology Laboratory, The Salk Institute.[d]
Supported by National Institutes of Health Grant Nos. RO1-HD-18967, P50-HD-12629, and P01-HD-13527, and conducted in part by the University of Washington Clinical Research Center (Grant No. RR-37), the Veterans Administration, and the Clayton Foundation for Research, California Division.
Presented by invitation at the Ninth Annual Meeting of the American Gynecological and Obstetrical Society, Hot Springs, Virginia, September 6-8, 1990.
W.W.V. is a senior Clayton Foundation investigator.
Reprint requests: Michael R. Soules, MD, Department of Obstetrics and Gynecology, RH-20, University of Washington, Seattle, WA 98195.
6/6/27369

There is a complex relationship between the hypothalamic-pituitary (neuroendocrine) unit and the corpus luteum in women. A well-described shift in luteinizing hormone (LH) pulse frequency and amplitude occurs from the follicular to the luteal phase, with fewer LH pulses of higher amplitude present in the second half of the cycle. This slowing of the LH secretion pattern in the luteal phase is a result of progesterone feedback on the hypothalamic gonadotropin-releasing hormone (GnRH) pulse generator, which is mediated by endogenous opioid peptides.[1, 2] Progesterone secretion by the corpus luteum is pulsatile as well and has been shown to be coupled with LH pulses in the midluteal

and late luteal phases.[3-5] LH appears to be required in both a permissive and a quantitative manner for normal progesterone secretion by the corpus luteum.[3, 6] Because the follicular phase LH pulse pattern is of relatively high frequency and low amplitude, there is a necessary increase in the LH secretory rate that accompanies the death of the corpus luteum. Withdrawal of progesterone is the presumptive stimulus for the luteal to follicular increase in gonadotropin pulse frequency.

Luteal phase deficiency is a well-described aberration of the menstrual cycle that usually consists of decreased progesterone secretion by the corpus luteum. Luteal phase deficiency can be a phenomenon that occurs sporadically in normal women, without clinical significance, or it can be the cause of infertility or habitual abortion.[7] We have reported significant abnormalities in the hypothalamic-pituitary-ovarian axis in women with luteal phase deficiency. They have an increased LH pulse frequency in the early follicular phase, decreased mean LH levels during the LH surge, decreased bioactive LH levels in the midluteal and late luteal phases, and decreased progesterone pulse amplitude in the luteal phase, accounting for their low integrated progesterone levels.[8, 9] In a separate study we were able to produce luteal phase deficiency in normal women by inducing a supraphysiologic LH pulse frequency during the follicular phase.[10] On the basis of these studies, we consider the rapid early follicular LH pulse pattern a key element in the pathophysiology of luteal phase deficiency and believe it is the probable cause of the abnormal LH and progesterone secretion later in the cycle.

Thus with our focus on the rapid early follicular LH pulse pattern as a key element in the pathogenesis of luteal phase deficiency and the ability of progesterone to modulate the LH pulse frequency, we hypothesized that luteal phase deficiency could be self-perpetuating. The following hypothesis was entertained: decreased progesterone secretion in a given luteal phase could result in an excessively rapid gonadotropin pulse pattern in the subsequent follicular phase which, in turn, would lead to luteal phase deficiency in that and future cycles. This study was designed to test this hypothesis. To this end, luteal phase deficiency (premature luteolysis with decreased luteal progesterone levels) was induced in normal women with a GnRH antagonist and the subsequent cycle was monitored in terms of the follicular LH secretion pattern and corpus luteum function.

Methods

Subjects. This study was designed for normal volunteers to serve as their own controls. These women ($n = 6$) were between 26 to 33 years of age, within ±10% of their ideal body weight (Metropolitan Life Insurance Co. tables, 1980), and no more than moderate exercisers. They had a history of 27- to 32-day menstrual cycles, at least one normal biphasic basal body temperature chart, and met the following hormonal criteria in the midluteal phase of a menstrual cycle preceding the study: progesterone ≥12 ng/ml, prolactin <20 ng/ml, and testosterone ≤40 ng/dl. The protocol was approved by the Human Subjects Committee of the University of Washington and informed consent was obtained from each woman.

There was a preset criterion for inclusion in the study that the GnRH antagonist administration induce a luteolysis sufficiently similar in terms of the pattern and time course of decreased progesterone levels to that seen in women with spontaneous luteal phase deficiency as previously described.[8] To this end, we required each subject to have a luteal phase of 13 or more days and an integrated progesterone level >100 ng/ml per day in the control cycle. In the luteal phase of the treated cycle they had to have an integrated progesterone level <100 ng/ml per day that was also a minimum of 33% below the integrated progesterone in their control cycle. Whereas a more profound and dramatic luteolysis could have been achieved with the Nal-Glu GnRH antagonist, depending on the dose, the decreased progesterone levels observed in spontaneous luteal phase deficiency cycles tend to be modest (around 33%) and usually occur in the latter half of the luteal phase. Some women with luteal phase deficiency had entirely normal levels of progesterone until the midluteal phase.[8] Fifteen subjects volunteered for the study and six met the criteria. Of the subjects who did not meet the study criteria, five had low integrated progesterone levels in the control cycle and the other four had insufficient luteolysis.

Protocol. The study encompassed three menstrual cycles. The protocol is summarized in Fig. 1. Daily blood samples were obtained from cycle day 10 until the onset of the next menstrual period in all three cycles. These samples were allowed to clot, serum was separated, aliquoted, and frozen at −4° C until assayed for LH and progesterone. A 12-hour admission to the Clinical Research Center occurred in the early follicular phase (cycle days 1 to 4) of the second and third study cycles. During these admissions (8 AM to 8 PM) a heparinized intravenous line was placed, through which 5 ml venous blood samples were drawn every 10 minutes (total, 73 samples). The serum from these samples was frozen at −4° C and subsequently assayed for LH.

The Nal-Glu antagonist injections were initiated in the midluteal phase of study cycle two and were administered subcutaneously every 12 hours. The injections began on cycle days +5 or +6 in relation to the LH surge (day 0) and were continued until the onset of the next menstrual period. The dose of the Nal-Glu

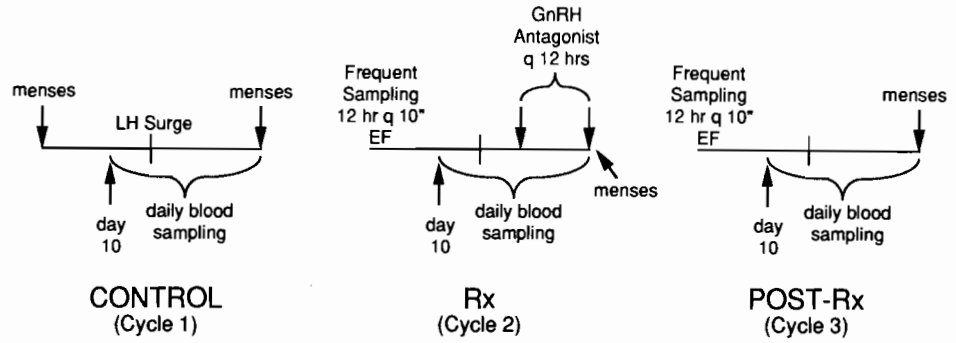

Fig. 1. The study protocol encompassed three menstrual cycles. The luteal phase of cycle one and the early follicular *(EF)* phase of cycle two were control intervals for luteal progesterone and follicular LH secretion, respectively. Luteal phase deficiency was induced with a GnRH antagonist administered from the midluteal phase of cycle two until the subject's next menstrual period (*Rx*, treatment). LH and progesterone secretion were monitored in cycle three.

antagonist was determined on the basis of our previous experience with its administration in the luteal phase[11] and was either 2.5 μg/kg or 5.0 μg/kg for each injection. The daily blood sampling increased to every 12 hours during antagonist administration, which varied between 5 and 8 days in duration.

Assays. Serum LH was measured by double antibody radioimmunoassay as previously described[12] with reagents supplied by the National Institutes of Health with LER 907 as the reference preparation. The sensitivity of the LH assay was 6 ng/ml, with intrassay and interassay coefficients of variation of 5.5% and 8.4%, respectively.

Serum progesterone was measured by radioimmunoassay with reagents supplied by Diagnostic Products Corp. (Los Angeles). This assay had a cross-reactivity of 0.3% with 17-hydroxyprogesterone and <0.01% with testosterone, estradiol, pregnenolone, and cortisol. The assay sensitivity was 0.5 ng/ml, and intraassay and interassay coefficients of variation were 9% and 12%, respectively.

Statistics. Luteal phase length in this study was defined as the number of days that occurred subsequent to the LH surge (peak LH value, day 0) and before the next menstrual period. Integrated progesterone values were calculated for each subject's luteal phase by adding the daily serum progesterone levels together beginning with the day of the LH surge; they are expressed as nanograms per milliliter per day.

For each subject there were 73 LH data points from the early follicular phase admissions in study cycles two and three. The following pulse parameters were calculated for LH: frequency, amplitude, and mean level. An adaptive threshold method (DC3) was used to determine the frequency and amplitude of pulses as previously described.[13]

Each volunteer served as her own control. Comparisons were made between study cycles with the paired Student t test for luteal length, integrated progesterone, and pulse parameters. The data are presented as mean ± SEM unless otherwise indicated. A p value ≤ 0.05 was considered significant.

Results

A decrease in serum progesterone levels in cycle two that simulated luteal phase deficiency was a requirement of this study. Thus mean integrated progesterone levels over the luteal phase decreased from 136.9 ± 12.9 to 80.7 ± 7.2 ng/ml per day between the control and treatment cycles (Fig. 2). Not unexpectedly, mean luteal phase length also decreased from 13.7 ± 0.6 to 11.2 ± 0.8 days ($p < 0.01$) between the control and treated cycles (Fig. 2). Therefore a luteal phase deficiency–like condition was successfully induced in these normal women.

The posttreatment cycle was carefully examined to ascertain whether this luteal phase deficiency state had a carry-over effect(s) into the subsequent cycle. The LH pulse pattern in the early follicular phase of the posttreatment cycle was compared with the pattern before treatment. A representative pair of early follicular LH secretion patterns from a particular volunteer are illustrated in Fig. 3. Both patterns appear to represent normal early follicular LH secretion. In fact, when the LH secretory parameters were compared for the entire group, there were no differences in frequency, amplitude, or mean level between the control and posttreatment sampling intervals (Table I).

In terms of luteal phase length and serum progesterone levels, the posttreatment cycle was similar to the control cycle. There was a tendency toward a decreased mean luteal phase length (12.7 ± 0.6 vs 13.7 ± 0.6 days, respectively, $p = 0.08$, Fig. 2). However, the mean integrated serum progesterone levels were nearly iden-

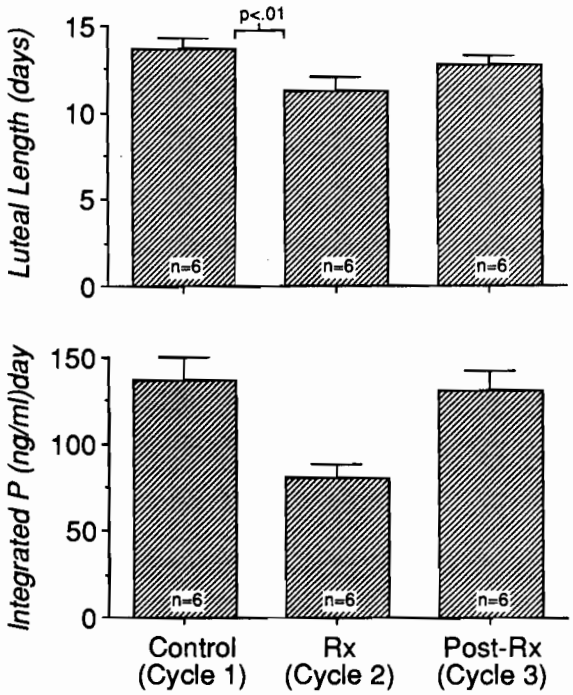

Fig. 2. Corpus luteum function in each of the three cycles is illustrated. Luteal phase deficiency was successfully induced in the treatment *(Rx)* cycle.

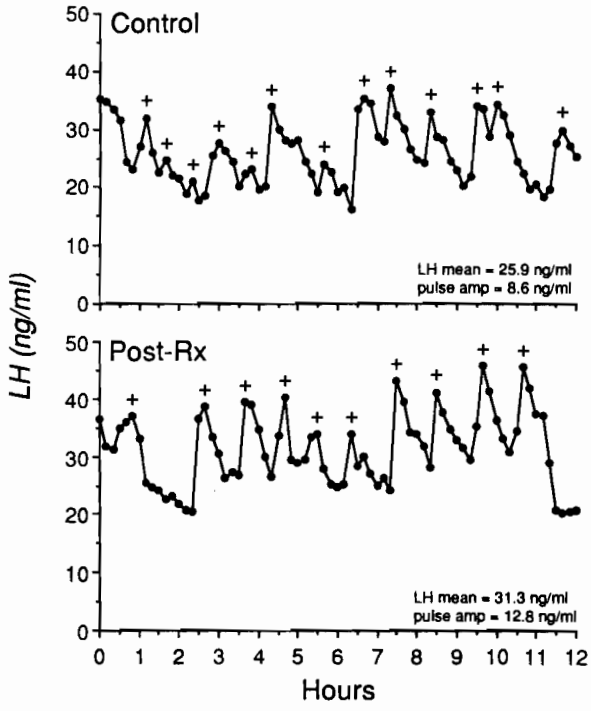

Fig. 3. The LH secretory pattern over 12 hours is illustrated for subject B in the early follicular phase of the control and posttreatment cycles. Secretory episodes *(pulses)* are indicated with a *plus sign.*

tical at 130.5 ± 11.3 vs 136.9 ± 12.9 ng/ml per day, $p = 0.5$ (Fig. 2). Therefore there was no evidence of a luteal phase deficiency–like condition in the posttreatment cycle.

Comment

Luteal phase deficiency is both a controversial and a diverse disorder with a number of investigators debating whether it actually occurs. It certainly does exist because it has been well documented in many different settings: post menarchal[14]; premenopausal[15]; post partum[16]; with strenuous exercise[17]; with dieting[18]; and in "normal" women who volunteer as control subjects.[19, 20] It can be induced by interfering with folliculogenesis,[21] luteal phase luteinizing hormone suppression,[22] or a supraphysiologic early follicular LH pulse frequency.[10] Therefore the relevant question is not whether it exists, but does it occur regularly in some women and thereby cause infertility or recurrent abortions? The latter question remains controversial, primarily because there is not a simple effective test to determine whether luteal phase deficiency is present in the clinical setting.[23] It is our considered opinion that luteal phase deficiency is a primary cause of infertility and habitual abortion in 5% to 10% of a population referred for subspecialty care.

Luteal phase deficiency consists of either decreased corpus luteum function (inadequate secretion of progesterone, estradiol, and inhibin) or an insufficient hormonal effect at the level of the endometrium. Whereas considerable research attention has been directed at insufficient end-organ response in luteal phase deficiency, very little consistent evidence has come forth.[23] Therefore in both the research and clinical setting, luteal phase deficiency as we understand it today can primarily be characterized as inadequate hormonal secretion. Many reasons for inadequate hormonal secretion by the corpus luteum have been proposed: (1) inadequate follicular development, (2) a dysfunctional LH surge, (3) insufficient LH support of the corpus luteum in the luteal phase, (4) endogenous corpus luteum defect(s) that preclude adequate function, and (5) premature or excessive luteolysis.

There has been a relative paucity of studies that have investigated the pathogenesis of spontaneous luteal phase deficiency in women, especially with regard to endogenous defects in the corpus luteum itself. There are good indications that interference with folliculogenesis can lead to luteal phase deficiency[21] and abnormal follicular phase follicle-stimulating hormone (FSH) levels have been reported in luteal phase deficiency.[19, 20, 24] However, we were not able to find any abnormalities in FSH levels, follicle size, or estradiol levels in an in-depth study of 10 women with sponta-

Table I. LH secretory parameters in the early follicular phase

Cycle	*Subject*	*LH pulses (No. per 12 hr)**	*LH pulse amplitude (ng/ml)†*	*Mean LH (ng/ml)‡*
Control				
	A	9	12.9	25.3
	B	13	8.7	26.0
	C	6	14.8	20.4
	D	10	5.8	11.4
	E	8	11.0	15.1
	F	11	12.6	18.5
	Mean ± SEM	9.5 ± 1.0	11.0 ± 1.3	19.4 ± 2.3
Posttreatment				
	A	10	16.1	31.3
	B	10	12.8	31.3
	C	6	20.1	24.9
	D	12	8.2	15.9
	E	10	7.2	13.7
	F	12	7.3	16.0
	Mean ± SEM	10.0 ± 0.9	12.0 ± 2.2	22.2 ± 3.3

*$p = 0.5$.
†$p = 0.6$.
‡$p = 0.1$.

neous luteal phase deficiency.[8] That study did find deficiencies in the LH surge and in the LH support of the corpus luteum. We believe this lack of hormonal stimulus to the corpus luteum is a major component of the pathophysiology of luteal phase deficiency. In addition, two further pieces of evidence led us to focus on the neuroendocrine component (GnRH pulse generator) as the basis for luteal phase deficiency in most women with this condition. First, we have quite consistently found a significant increase in LH pulse frequency (with the expected decrease in LH pulse amplitude) in the early follicular phase of luteal phase deficiency cycles.[9] Other investigators have found abnormalities (both rapid and slow) in early follicular LH pulse frequency in luteal phase deficiency as well.[25, 26] Second, we were able to induce significantly decreased luteal phase levels of progesterone and bioactive LH in normal women by means of a supraphysiologic LH pulse frequency in the follicular phase.[10] Therefore it appeared to us that investigating the altered GnRH pulse generator in the follicular phase might be the key to understanding the pathophysiology of luteal phase deficiency.

One possible cause of the increased secretory rate of LH in luteal phase deficiency could be the low progesterone levels in the prior luteal phase. Progesterone is known to modulate (slow) the GnRH pulse generator,[1] thus too little progesterone could lead to a premature and excessive increase in LH secretion and, potentially, luteal phase deficiency in the next cycle. This study found no compelling evidence that luteal phase deficiency perpetuates itself into subsequent cycles. The lack of decreased progesterone secretion was to be expected after induced luteal phase deficiency when no changes in the LH secretory pattern in the early follicular phase occurred. It now becomes necessary to look at other modulators of the GnRH pulse generator to understand the alterations in early follicular LH secretion and the pathophysiology of luteal phase deficiency. There are many examples of suprahypothalamic influences on the GnRH pulse generator that have profound effects on the menstrual cycle. Conditions such as anorexia nervosa,[27] hypothalamic amenorrhea,[28] heavy exercise,[29] and dieting[30] all have in common either psychological or physical stress, abnormal gonadotropin secretion patterns, and menstrual dysfunction. We postulate that luteal phase deficiency as a mild and subtle form of menstrual dysfunction commonly occurs because of stress-induced alterations in the GnRH pulse generator, mediated by neurotransmitters.

We gratefully acknowledge the laboratory assistance of Dorothy McGuinness, Florida Flor, and Arlen Sarkissians; the illustration assistance of Jan Hamanishi and Patrick Clarke; the data analysis of Dawn McCracken; and the editorial assistance of Nancy Cohen.

REFERENCES

1. Soules MR, Steiner RA, Clifton DK, Cohen NL, Aksel S, Bremner WJ. Progesterone modulation of pulsatile luteinizing hormone secretion in normal women. J Clin Endocrinol Metab 1984;58:378-83.
2. Quigley ME, Yen SS. The role of endogenous opiates on LH secretion during the menstrual cycle. J Clin Endocrinol Metab 1980;51:179-81.
3. Soules MR, Clifton DK, Steiner RA, Cohen NL, Bremner WJ. The corpus luteum: determinants of progesterone secretion in the normal menstrual cycle. Obstet Gynecol 1988;71:659-66.
4. Filicori M, Butler JP, Crowley WF. Neuroendocrine regulation of the corpus luteum in the human: evidence for

pulsatile progesterone secretion. J Clin Invest 1984;73: 1638-47.
5. Veldhuis JD, Christiansen E, Evans WS, Kolp LA, Rogol AD, Johnson ML. Physiological profiles of episodic progesterone release during the midluteal phase of the human menstrual cycle: analysis of circadian and ultradian rhythms, discrete pulse properties, and correlations with simultaneous luteinizing hormone release. J Clin Endocrinol Metab 1988;66:414-21.
6. Cohen NL, Clifton DK, Bremner WJ, Dahl KD, Soules MR. Induced luteinizing hormone pulses exert a quantitative influence on progesterone secretion in normal women [Abstract]. In: Proceedings of the Society for Gynecologic Investigation 37th Annual Meeting. St. Louis: Society for Gynecologic Investigation, March 1990.
7. Soules MR. Luteal phase deficiency: an underdiagnosed and overtreated reproductive endocrine disorder. Obstet Gynaecol Clin North Am 1987;14:865-85.
8. Soules MR, McLachlan RI, Ek M, Dahl KD, Cohen NL, Bremner WJ. Luteal phase deficiency: characterization of reproductive hormones over the menstrual cycle. J Clin Endocrinol Metab 1989;69:804-12.
9. Soules MR, Clifton DK, Cohen NL, Bremner WJ, Steiner RA. Luteal phase deficiency: abnormal gonadotropin and progesterone secretion patterns. J Clin Endocrinol Metab 1989;69:813-20.
10. Soules MR, Clifton DK, Bremner WJ, Steiner RA. Corpus luteum insufficiency induced by a rapid gonadotropin-releasing hormone-induced gonadotropin secretion pattern in the follicular phase. J Clin Endocrinol Metab 1987;65:457-64.
11. McLachlan RI, Cohen NL, Vale WW, et al. The importance of luteinizing hormone in the control of inhibin and progesterone secretion by the human corpus luteum. J Clin Endocrinol Metab 1989;68:1078-85.
12. Midgley AR. Radioimmunoassay: a method for human chorionic gonadotropin and human luteinizing hormone. Endocrinology 1966;79:10.
13. Clifton DK, Aksel S, Bremner WJ, Steiner RA, Soules MR. Statistical evaluation of coincident prolactin and luteinizing hormone pulses during the normal menstrual cycle. J Clin Endocrinol Metab 1988;67:832-8.
14. Apter D, Vihko R. Early menarche, a risk factor for breast cancer, indicates early onset of ovulatory cycles. J Clin Endocrinol Metab 1983;57:82-6.
15. Lenton EA, Landgren BM, Sexton L. Normal variation in the length of the luteal phase of the menstrual cycle: identification of the short luteal phase. Br J Obstet Gynaecol 1984;91:685-9.
16. Gray RH, Campbell OM, Zacur HA, et al. Postpartum return of ovarian activity in nonbreast feeding women monitored by urinary assays. J Clin Endocrinol Metab 1987;64:645-50.
17. Bullen BA, Skrinar GS, Beitins IZ, et al. Induction of menstrual disorders by strenuous exercise in untrained women. N Engl J Med 1985;312:1349-53.
18. Pirke KM, Schweiger U, Lemmel W, Krieg JC, Berger M. The influence of dieting on the menstrual cycle of healthy young women. J Clin Endocrinol Metab 1985;60:1174-9.
19. Strott CA, Cargille CM, Ross GT, et al. The short luteal phase. J Clin Endocrinol Metab 1970;30:246-51.
20. Sherman BM, Korenmann SG. Measurement of plasma LH, FSH, estradiol and progesterone in disorders of the human menstrual cycle: the short luteal phase. J Clin Endocrinol Metab 1974;38:89-93.
21. Stouffer RL, Hodgen GD. Induction of luteal phase defects in rhesus monkeys by follicular fluid administration at the onset of the menstrual cycle. J Clin Endocrinol Metab 1980;51:669-71.
22. Mais V, Kazer RR, Cetl NS, Rivier J, Vale W, Yen SS. The dependency of folliculogenesis and corpus luteum function on pulsatile gonadotropin secretion in cycling women using a gonadotropin-releasing hormone antagonist probe. J Clin Endocrinol Metab 1986;62:1250-5.
23. McNeely MJ, Soules MR. The diagnosis of luteal phase deficiency: a critical review. Fertil Steril 1988;50:1-15.
24. Cook CL, Rao CW, Yussman MA. Plasma gonadotropin and sex steroid hormone levels during early, midfollicular and midluteal phases of women with luteal phase defects. Fertil Steril 1983;40:45-8.
25. Suh BY, Betz G. Altered frequency of LH secretion in luteal phase defects in patients (LPD) [Abstract]. In: Proceedings of the Society for Gynecologic Investigation 35th annual meeting. Baltimore: Society for Gynecologic Investigation, March 1988.
26. Schweiger U, Laessle RG, Tusehl RJ, Broocks A, Krusche T, Pirke KM. Decreased follicular phase gonadotropin secretion is associated with impaired estradiol and progesterone secretion during the follicular and luteal phases in normally menstruating women. J Clin Endocrinol Metab 1989;68:888-92.
27. Yen SS. Chronic anovulation due to CNS-hypothalamic pituitary dysfunction. In: Yen SS, Jaffee RB, eds. Reproductive endocrinology. Philadelphia: Saunders Press, 1986:510.
28. Reame NE, Sauder SE, Case GD, et al. Pulsatile gonadotropin secretion in women with hypothalamic amenorrhea: evidence that reduced frequency of gonadotropin-releasing hormone secretion is the mechanism of persistent anovulation. J Clin Endocrinol Metab 1985;61:851.
29. Veldhuis JD, Evans WS, Demers LM, Thorner MO, Wakat D, Rogol AD. Altered neuroendocrine regulation of gonadotropin secretion in women distance runners. J Clin Endocrinol Metab 1985;61:557-63.
30. Cameron JL, Nosbisch C. Slowing of pulsatile LH and testosterone secretion during short-term fasting in adult male rhesus monkeys *(Macaca mulatta)* [Abstract]. Endocrinology [In press].

Discussion

Dr. Isaac Schiff, Boston, Massachusetts. Dr. Soules is clearly a leader in the field of luteal phase deficiency. His solid and important research has furthered our knowledge of the subject since Dr. Georgeanna Seegar Jones first described the condition over 40 years ago. Luteal phase deficiency was defined as the unusual condition in which the endometrium was greater than 2 days out of phase as documented by endometrial sampling. The treatment that was suggested was progesterone supplementation. Numerous studies that indicate the beneficial results of progesterone therapy for luteal phase deficiency, when traditionally diagnosed with a biopsy specimen, constitute provocative evidence for this disorder being a cause of reproductive failure.

I read Dr. Soules' article with great interest and in the brief time allowed me here I would like to formulate my comments on it in a number of questions—it being true that every scientific article of significance raises as many questions as it answers.

His protocol asked whether normal women can withstand an assault on their hypothalamic pituitary gonadal axis by internally adapting and resetting the pulse generator. The answer to the posed question was clearly no. There was no evidence of induction of a luteal phase deficiency–like condition. Perhaps some persons are more susceptible to hypothalamic pituitary gonadal axis perturbations than are others. Is it true that most persons with some stress or external stimuli can adapt and reorganize the hypothalamic pituitary gonadal axis

for the next menstrual cycle and therefore resist the tendency of luteal phase deficiency to develop? Is it true that others cannot because of some defect of the hypothalamic pituitary gonadal axis that, although subtle, is serious enough to produce luteal phase deficiency? One could thus obtain different data if more patients were studied or if an infertile group of women were so examined. After all, the authors state that luteal phase deficiency can be a phenomenon that occurs sporadically in normal women. On this same matter, how long do the effects of alteration in progesterone persist to control gonadotropin secretion?

In his article, Dr. Soules stresses the LH pulsations as an explanation for luteal phase deficiency. Given the unfair advantage of hindsight and being cognizant of the negative results, one might ask whether there is too much emphasis on LH. According to the work of Hutchison, the corpus luteum can be maintained with infrequent pulses of GnRH (hence LH) during the luteal phase, but its usual duration cannot be extended with physiologic doses. And indeed other factors such as prostaglandins may be operative. High levels of LH in the follicular phase may induce multiple folliculogenesis, and the increase in estrogens from these follicles could produce premature luteinization and the corpus luteum defect. Could, then, the addition of LH in the luteal phase prevent luteal phase deficiency? Finally, if LH is that critical then it implies that the hypothalamus pituitary-luteal axis is homeostatic, as are other pituitary target organs. I think that this is not the case, and it is a good thing too, because then the pituitary would always be trying to rescue the corpus luteum at the end of each nonconception cycle.

Some authors believe that luteal phase deficiency might be induced by an alteration in FSH levels in the follicular phase. Dr. Soules, did you happen to measure FSH, and if so, did you find a difference?

In the day-to-day clinical situation, many physicians do not have access to Dr. Soules' laboratory expertise, and they are compelled to rely on results of endometrial biopsies. Given their well-known drawbacks, I wonder whether in carrying out his study Dr. Soules performed any biopsies?

To induce experimentally the condition of corpus luteum defect, the investigators used a GnRH antagonist. The procedure, as the authors acknowledge, shortens the luteal phase. The short luteal phase might itself be only one type of corpus luteum defect. A further reaching study design would have called for a luteal phase of 12 to 14 days' duration and an integrated progesterone level that is decreased. I welcome Dr. Soules' suggestions on this point.

Perhaps there is another conceptual framework for the model of the self-perpetuating nature of luteal phase deficiency. If we assume that the following statements are more or less basic observations: (1) progesterone in nonconception cycles declines at some point and (2) the pulse generator does not count the days from the LH surge and it does not act differently when luteolysis occurs early, inasmuch as it always has to recover from a fall in progesterone to renew the state of estrus, I wonder whether Dr. Soules would comment about a classic homeostatically oriented hypothesis that would theorize that follicular recruitment and subsequent luteal function would if anything be more robust after a short, insufficient luteal phase.

In summary, Dr. Soules has achieved remarkable results in the delineation of luteal phase deficiency. His careful, meticulous studies led him to reason that decreased progesterone in the luteal phase might alter LH pulsatile secretion in the subsequent follicular phase. The idea constituted a brilliant attempt to consolidate a pathophysiologic vision about a difficult question in a single study. Unfortunately, this did not turn out to be the case. Therefore decreased progesterone production in one cycle does not lead to recurrent luteal phase defects in normal women, implying important evidence that supports the complex cause of this syndrome. Thus we will await with great expectations his follow-up studies that may shed further light on the matter.

REFERENCES

1. Filicori M. Maintenance of the corpus luteum of the menstrual cycle: hypothalamo-pituitary-ovarian axis. Semin Reprod Endocrinol 1990;8:115-21.
2. Auletta FJ, Schofield MJ, Abae M. The mechanisms controlling luteolysis in non-human primates and women. Semin Reprod Endocrinol 1990;8:122-9.
3. Gibson M. Clinical evaluation of luteal function. Semin Reprod Endocrinol 1990;8:130-41.

Dr. James Schreiber, Chicago, Illinois. The hypothesis was that low progesterone would set off the next cycle to make it short. I was curious whether another way to approach this would be to use an antiprogestin such as RU 486, mifepristone, and block progesterone directly rather than perturbing centrally with the GnRH antagonist.

Dr. Daniel H. Riddick, Burlington, Vermont. I wonder whether you have looked at the converse of your experimental design. Have you taken women with luteal phase deficiency and supplemented the progesterone to normal levels in a deficient cycle and then studied the subsequent cycle with no treatment in those women?

Dr. Soules (Closing). I thank Dr. Schiff for a very thorough review of the article and his thoughtful questions.

The first question was in regard to the time course of effect of altering progesterone on LH secretion. The LH pulse generator (the gonadotropin generator) accelerates when progesterone is not present. On the basis of our studies and the literature, when progesterone levels decrease in the midluteal to late luteal phase, the LH pulse generator speeds up. The increase in the LH secretory takes place over 2 to 4 days.

He brought up a good point: we did not necessarily prove the null hypothesis by studying only six women. We could have studied more women and found that our hypothesis was actually true or we could have stud-

ied more susceptible women (e.g., infertile women) and may have found that our hypothesis was true. That is a good criticism. However, I happen to doubt that the study of more or different subjects would have changed our findings because we did not even see a trend in our data toward luteal phase deficiency in the menstrual cycles after induced luteolysis.

Dr. Schiff asked whether addition of LH in the luteal phase could prevent luteal phase deficiency. Yes, I believe it can. In fact, human chorionic gonadotropin supplementation is an acceptable therapy for luteal phase deficiency. It is not used very often; but in my experience, human chorionic gonadotropin does work.

The fourth question was provocative in regard to the pituitary and whether it had a homeostatic relationship with the ovary. His example was the pituitary-thyroid axis wherein the pituitary does everything it can to keep the end organ functioning when it fails. I think the pituitary ovarian axis functions in the same manner. In the late luteal phase when there is the normal decrease in progesterone, the gonadotropin pulse generator accelerates with a corresponding increase in the mean levels of LH and FSH. The hypothalamic pituitary unit is trying to rescue the corpus luteum, but the corpus luteum remains resistant. Therefore the pituitary does act in its usual homeostatic manner with the ovary as well.

The next question was whether we measured FSH in these particular patients. The answer is no, but I would like to allude to our 1989 study in which we intensively studied 10 women with spontaneous luteal phase deficiency. In that study, we looked at FSH every way we could to determine whether there were any changes in FSH. We looked at immunoactive FSH, bioactive FSH, follicular size, and estradiol levels. In these infertile women with spontaneous luteal phase deficiency, we could not find any evidence that abnormalities in FSH or follicular development were a part of the pathogenesis of luteal phase deficiency. However, it is clear in the literature that altering FSH or follicular development can cause luteal phase defect.

The next question was with regard to biospies. No, we did not obtain endometrial biopsy specimens because we think the integrated progesterone level provides a much more accurate diagnosis of luteal phase deficiency.

Dr. Schiff's next question was whether we could have induced a different type of luteal phase deficiency (low progesterone levels but with normal luteal length). This is the most common pattern we see in spontaneous luteal phase deficiency. It would have been nice to induce this pattern, but our experience with the Nal-Glu GnRH antagonist is that you can not be that precise. In fact, we experienced the opposite problem of luteolysis being induced too quickly, wherein we dropped the bottom out of the corpus luteum immediately in some dose studies that preceded this study. So I think more prolonged luteolysis would have been a nice study design, but this was beyond the capabilities of our study drug.

The next question concerned our hypothesis. Dr. Schiff theorized that if you suppressed the corpus luteum, you could give the follicular phase more time to develop and may see a more robust ovarian response in the next cycle. That is an interesting alternative hypothesis. The natural response of the hypothalamic-pituitary unit to decreased levels of progesterone is to raise circulating levels of LH and FSH that promote follicular development in the ensuing cycle. In formulating our hypothesis we reasoned that if the hypothalamus were exposed to less progesterone over a shorter period of time, we could find a supraphysiologic acceleration in the LH secretion pattern. In our research experience wherein we induced a supraphysiologic LH secretion pattern we induced luteal phase deficiency. Furthermore, I am not aware of a natural (physiologic) gonadotropin secretion pattern that leads to healthier, more robust follicles. We expected to find either inadequate or normal follicular development in response to luteolysis and we found the latter. It would have been fascinating to have found a more robust subsequent cycle but that was a less plausible hypothesis based on our knowledge of ovarian physiology and our research experience.

Dr. Schreiber asked about a different study design, wherein the effect on the next cycle could be examined after a partially blocking progesterone feedback effect on the hypothalamic-pituitary unit with the progesterone antagonist RU 486. At one time, we considered the use of RU 486 for this purpose but decided against it because we could think of no way to monitor whether we induced a complete or partial block. To mimic luteal phase deficiency only a partial progesterone block at the hypothalamus-pituitary is necessary.

Finally, Dr. Riddick asked whether we have treated women with luteal phase deficiency with progesterone to determine whether the pulse pattern is normal in the subsequent follicular phase. This proposed study is a logical corollary to the study I just presented and we plan to start the very study Dr. Riddick has proposed within the next year.