J Clin Endocrin Metab. First published ahead of print July 22, 2008 as doi:10.1210/jc.2008-0482

Follicle stimulating hormone and its rate of change in defining menopause transition stages

Abbreviated title: FSH change and menopause transition stages

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DISCLOSURE STATEMENT: The authors have nothing to disclose.

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Word Count: 3,723

Key Terms: stages of reproductive senescence, menopause staging, FSH, hormone trajectories, ovarian aging

Grants supporting the writing of this manuscript: AR051384 (Sowers, PI), AR040888 (Sowers, PI),

AR-20557 (Sowers, PI)

ABSTRACT (word count=269)

Context/Objective: To identify menopause transition stages using acceleration or deceleration patterns of follicle stimulating hormone (FSH) rates of change from the late reproductive years to postmenopause.

Setting/Participants: Michigan Bone Health and Metabolism Study cohort of 629 women, aged 24-44 years (in 1992/3), with 5757 annual FSH data points over a 14-year period.

Design/Main Outcome Measures: Relate acceleration/deceleration patterns in FSH rate of change to time to final menstrual period (FMP) and chronological age using non-parametric and piecewise regression modeling.

Results: Four major FSH stages, based on rate of FSH change patterns, were identifiable in relation to the FMP. In FSH stage 1, the rate of FSH change increased modestly up to -7 years prior to the FMP; in FSH stage 2 (-7 to -2 years prior to FMP), there was a major acceleration in FSH rate of change. FSH stage 3 had an acute increase in FSH rate of change (-2 to +1 years around the FMP), with average FSH level of 34 mIU/ml. The fourth, or plateau, FSH stage began at 1 year post-FMP when the average FSH level was 54 mIU/ml. During the years 28-60 years, there were eight epochs defined by significant changes of FSH trajectory accelerations or decelerations and rate of change.

Conclusions: Four menopause transition stages bounding the FMP and eight epochs in chronological aging from age 28 to 60 years were defined by changes of FSH trajectory accelerations/decelerations and rates of change. This timing information, combined with knowledge of FSH levels and menstrual cycle characteristics, can help discern the likely status of women with respect to their reproductive viability and menopause transition stage.

INTRODUCTION

More information about the follicle-stimulating hormone (FSH) patterns across the reproductive period and through the menopause transition is needed to help refine definitions of increasingly diminished ovarian reserve (representing the quantity and quality of the ovarian follicle pool) (1,2) and the transition stages of the menopause period. This need for information was made prominent by the STRAW report where there was a call to incorporate biomarker data, especially FSH information, with menstrual bleeding information to more adequately characterize the reproductive stages to the postmenopause (3).

Intracycle and intercyle variations in FSH (4,5) and the inhibins (6) have been well-characterized and are associated with the selection of a dominant follicle in the ovulatory process (7-9). However, current reports of FSH levels are limited in their ability to describe the natural history of both diminished ovarian reserve and the stages of the menopause (10). Though Burger et al. (11) was among the first to use longitudinally-acquired data to describe FSH levels in relation to chronological age and the final menstrual period (FMP), these investigators reported data from women who were aged 45 to 55 at their first annual examination, precluding their ability to describe women in their late reproductive years with increasingly diminished ovarian reserve and adequately link this information to define stages of the menopause transition.

This report addresses the natural history of change in FSH levels through the late reproductive years and into the menopause stages using FSH data from women who were aged 24-44 at their initial annual examination and who have been followed with annual assessments from 1992/3 through 2006/7. We characterized acceleration and deceleration of FSH rates of change, identifying patterns that could be aligned with reproductive senescence and menopause stages. We further described FSH patterns in relation to chronological age and time to final menstrual period (FMP), and then considered whether smoking behavior, parity, and age at menarche altered these patterns.

METHODS

Study population and sample size: The Michigan Bone Health and Metabolism Study (MBHMS) is a population-based longitudinal natural history study of reproductive hormones and their relation to the initiation and development of musculoskeletal and metabolic diseases and functional limitations (12,13). The study was implemented in a cohort of Caucasian women during their young and mid-adulthood. The 664-woman sample was identified from two sampling frames, the family records of Tecumseh (Michigan) Community Health Study from 1959-1985 and a 1992 Tecumseh community listing. In 1992, more 80% of the female Tecumseh offspring, aged 24-44 years, were recruited from the family records listing (n=543). Also recruited were n=121 (91%) of women listed in Kohl's Directory, aged 24-44 years, who had become Tecumseh community residents after completion of the health study. The baseline MBHMS study eligibility requirements were age (24-44 years), listing in the sampling frames, and sufficient mobility to attend a research clinic located in the community.

This report includes data from the 14-year period from 1992/3 through 2006/7. There were funding lapses in 1997 and 2003 for 18 and 14 months, respectively, during which neither data nor specimens were collected. For this report, 629 women contributed at least one data point to the 5,757 FSH data points available for the longitudinal data analyses. On average, participants contributed more than 9 annual FSH data points out of a total possible 11 annual points. Blood was not drawn (thereby precluding hormone analyses) if participants were pregnant or lactating at the time of the annual visit, recognizing that lactational amenorrhea is associated with overshoot in FSH levels (14). Over time, 2-7% of women had only interview data available at any given year because participants lived more than 2.5 hours from the research clinic, were too ill to contribute to an in-person visit with phlebotomy, or refused phlebotomy (<0.5%). Data were censored at time of death for the 14 (2%) participants who have died since the cohort inception.

This study was approved by the University of Michigan Institutional Review

Board and informed consent was obtained from all participants.

Measures of menopausal transition status: Menopause status was based on the regularity of menstrual bleeding in the year prior to the study visit. A woman was classified as premenopausal if she had no increase in menstrual irregularity in the previous year. Perimenopause was defined as having menstrual irregularity and having 9 or fewer menstrual cycles in a 12-month time period. Postmenopause was characterized as having at least twelve consecutive months of amenorrhea associated with no other medical cause. Final menstrual period (FMP) was defined retrospectively as 12 months of amenorrhea with no alternative physiologically normal explanation such pregnancy or lactation.

Surgical menopause, including hysterectomy and oophorectomy, was verified by medical record abstraction. Hormone therapy and oral contraceptive (HT/OC) use was assessed at each visit. Information included preparation components and duration of use. For purposes of these analyses, data from women with hysterectomy/oophorectomy were censored at the time of surgery and data at time of HT use were censored for that time point.

Sex steroid hormones: Blood and urine specimens were collected in a fasting state in days 2-7 of the follicular phase of the menstrual cycle. If a woman was postmenopausal or sufficiently advanced into the menopause transition that phlebotomy could not be linked to a menstrual bleed, specimens were collected on the anniversary of her study enrollment $\pm$ 15 days. Specimens were aliquoted and stored at -80 degrees Centigrade without thaw until assay. Serum Follicle Stimulating Hormone (FSH) concentrations were measured with a two-site chemiluminescence (sandwich) immunoassay which uses constant amounts of two antibodies that have a specificity for the intact FSH molecule(_1_); a polyclonal sheep anti-FSH antibody labeled with acridinium ester and a monoclonal mouse anti-FSH antibody covalently coupled to paramagnetic particles. Separation, aspiration, and deionized water wash steps separate bound from free. A direct relationship exists between the amount of FSH in the sample and the relative light units detected by the luminometer (photomultiplier tube). The test results are determined from a calibration curve using standards obtained from Bayer Diagnostics which are referenced to the WHO 2nd IRP 78/549 standard. The coefficients of variation (%) at locations along the standard curve (in parentheses) were as follows: 7.8% (3.3 mIU/ml), 3.2% (9.9 mIU/ml), 5.1% (18.2 mIU/ml), 4.4% (22 mIU/ml) and 3.3% (60.8 mIU/ml). The lower limit of detection was 1.05 mIU/mL.

Other measures: Height (cm) and weight (kg) were measured at each annual study visit with a stadiometer and balance-beam scale, respectively. Body mass index (BMI) was calculated by dividing the weight (in kilograms) by height (in meters) squared. Body composition was measured using bioelectrical impedance. Women were interviewed annually about selected aspects of their personal behavior and reproductive history. Smoking history and practice was ascertained annually. Participants were classified as never, past, current smokers. Parity was described based on the number of live births over 28 weeks of age. Age at menarche was self-reported.

Data analysis: Variable distributions were examined for normality, the presence of non-plausible outliers and/or changing variability over time. Univariate analyses were used to decide if transformations of outcome measures were necessary for satisfying model assumptions such as normality and constant variance. In analyses, FSH values were log transformed (natural) but back-transformed to facilitate ease of communication.

A multiple-step process, more fully described in Appendix I (Supplemental Data, published in *JCEM* online), was used to organize FSH rates of change into stage or epochs related to the final menstrual period (FMP) and chronological age. In the first step, because relationships between FSH values and time to final menstrual period (FMP) or chronological age could not be appropriately modeled by incorporating quadratic or cubic terms, a non-parametric stochastic mixed model was fit (15). 95% confidence intervals were fit around the mean FSH values in relation to time to FMP and chronological age. In the second step, to estimate the rate of change and

acceleration which are the first- and second-order derivatives of $_{log}$(FSH), respectively, we solved differential equations associated with the cubic spline function. The curvature of mean profile of $_{log}$FSH over time, representing the degree of bend in the line, was approximated by integrating both rate of change and acceleration/deceleration. The 95% confidence bands of these characteristics were obtained using bootstrapping approach with 100 bootstrap samples by replicating the above processes. In the third step, and informed by steps one and two, data were organized into epochs by setting nodes for piecewise linear mixed models using inflection points defined by differentiating the cubic spline smoothing functions (16). This estimated the mean rate of change at each segments in relation to time to FMP and chronological age. The population mean and standard errors for $_{log}$FSH at turning time points were presented in back-transformed scale for clinical understanding using Delta method Taylor series approach.

Analyses were implemented in SAS version 9.1 and SAS macro language (SAS Institute, Cary, NC), or Matlab7.0 (The MathWorks, Inc.).

RESULTS

At the 1992/3 baseline, the cohort median age was 38 (IQR = 7) while the median cohort age at the 2006/7 year examination was 51.9 years (IQR =7.3). The median baseline cohort BMI was 25.3 kg/m^2 (IQR = 7.4) and 29.4 kg/m^2 (IQR = 8.4) fourteen years later. The baseline median cohort fat mass was 23.8 kg (IQR =12.77) and 27.1 kg (IQR = 16.5) fourteen years later. The median skeletal muscle mass was 20.2 kg (IQR = 3.3) at baseline and 20.6 kg (IQR = 3.4) at the 2006/7 examination. The median age at menarche was 13 (IQR = 1) years and the median age at FMP was 50.5 years old.

Other characteristics of the population are shown in Table 1. Not surprisingly, over the time period, the number of women who were premenopausal (and not using exogenous hormones) declined from 70.6% at baseline to 23.8% at the 2006/7 visit; surgical menopause frequency changed from 4.4% of the cohort at baseline to 20.2% at the 2006/7 visit. Almost 15% of the cohort remained nulliparous across time and more than half of the cohort reported never smoking cigarettes.

FSH patterns in relation to the Final Menstrual Period (FMP). Figure 1 shows the rising mean population $_{log}$FSH levels in relation to the FMP with 95% confidence intervals (CI). Four different stages were identifiable in relation to time to the FMP using measures of acceleration and deceleration and piece-wise modeling of FSH rates of change (see Figure 2). Tables showing the regression beta coefficients and their 95% confidence intervals are shown in the Appendix.

FSH stage 1 was a period of gradually increasing FSH rate of change that ended 7 years prior to the FMP. At the node to FSH stage 2, there is significant acceleration to a greater FSH rate of change. This rate of change remained relatively constant during the time interval from -7 to -2 years prior to the FMP. During this period, the FSH levels increased, on average, from 15 mIU/ml to 33 mIU/ml. The node at FSH stage 3 marked an acute increase in the FSH rate of change; this increased rate was observed between -2 to +1 years around the FMP during which time the average FSH level rose from 34 mIU/ml to 54 mIU/ml. Finally, at the FSH stage 4 node and commencing 1 year post-FMP, there was downward shift in the FSH rate of change that resulted in a plateau of FSH levels. The mean chronological ages associated with the beginning of these stages were 43.6 years (Stage 2), 47.6 years (Stage 3), and 51 years (Stage 4).

The patterns of these stages, reflecting rate of FSH change, did not vary according to age at menarche, smoking status or parity.

Patterns of FSH with chronological age. Figure 3 shows the mean population values of FSH in relation to chronological age with 95% confidence intervals. Using non-parametric stochastic modeling and piece-wise linear mixed modeling**,** we identified eight epochs of accelerations and decelerations in the rates of FSH change over the chronological age period from 28 to 60. Each modeling approach identified similar patterns (see Table 3) as demonstrated by a relative difference in FSH levels of less than 3% between the two methods at matching time points. The eight distinct chronological age epochs in FSH rates of change

identified between ages 28 and 60 were 28-33, 33-40, 40-42, 42-45, 45-50, 50-52, 52-55, and 55-60 years.

The mean FSH (back-transformed) values at the critical age segments are shown in Table 3. A marked shift in the population mean FSH level is notable at the node for the age 45-50 segment where the mean FSH value at the beginning of the segment was 11.1 mIU/ml (SE=0.44). At the node for the age 50-52 segment, the mean FSH was 22.7 mIU/ml (SE=1.04) at the beginning of the segment; whereas, at the node for the age 52-55 segment, the mean FSH was 35.9 mIU/ml (SE=1.96). At the node for the age 55-60 segment, the mean FSH was 59.8 mIU/ml (SE=4.19) and, at the age 60 + segment, the mean FSH was 67.6 (SE=11.23), respectively.

Figure 4 shows the rates of change at different ages as well as the rates of acceleration or deceleration in those rates of change according to chronological age. The mean rates of change at each stage are shown in Table 2, Appendix. The age segments that were associated with statistically significant accelerations greater than zero ($P<0.05$) were observed at ages 40, 42, 45, and 50 years. After age 40, $_{log}$FSH increased rapidly and then achieved maximum rate of change between ages 50-52. At age 52, the FSH rate of change was more than 4 times faster than the rate of change observed in the stable period when women were in their mid-30s. After age 55, the rate of change in $_{log}$FSH was slightly positive but no longer statistically significant ($P=0.55$).

Women who continued to smoke or who had quit smoking had higher $_{log}$FSH at an earlier chronological age but not an increased rate of FSH change, even after adjusting for baseline age and baseline $_{log}$FSH. Neither parity nor age at menarche categorization was significantly associated with differential $_{log}$FSH levels or their rate of change at specific chronological ages.

DISCUSSION

This report characterizes the long-term natural history of FSH rate of change as a biomarker of the evolution from active reproduction to and through the menopause transition to the post-transition period. It also provides a contrast of FSH rates of change from the perspective of ovarian aging by referencing the final menstrual period (FMP) in comparison to rates of FSH change with respect chronological aging. In doing so, we identified four stages in FSH transition, based on acceleration or deceleration in the rate of FSH change in reference to the FMP. Current systems to describe the stages of the reproductive period and menopausal transition are based on variability in menstrual bleeding frequency (i.e., STRAW and ReStage) (3,17). Both systems have identified that it would be valuable to link menstrual bleeding criteria to biomarker levels but, to date, this has been hampered by the absence of longitudinal biomarker data that spans the reproductive and menopause transition time periods.

FSH has long been considered a candidate biomarker for describing reproductive and menopause transition stages (18) for two reasons. First, FSH stimulates folliculogenesis, a key in the dynamics of ovarian aging. Parallel with the age-related decline in the oocyte quality and quantity (19), there are progressively higher follicular phase FSH levels observed (20) in older ovulatory women compared to younger ovulatory women, believed to be due to the diminished restraint associated with declining inhibin-B levels, the dominant inhibin of the small antral follicles (21-24). Second, the rise in FSH levels precedes significant declines in ovarian steroid secretion, including that of estradiol (25).

Although the centrality of FSH to the ovulatory process has been long acknowledged and FSH cutpoints have been employed clinically to characterize subfertility as a criterion to initiate treatment, a clinical interpretation of actual FSH values in relation to menopause transition staging has been a source of concern. The well-known pulsatile, intracycle and intercycle variation in FSH levels make it difficult to interpret values without referencing biological landmarks including menstrual bleeding, ovulation or the LH surge. This measurement issue is coupled with the understanding that FSH levels are really an indirect indicator reflecting the diminishing restraint of inhibin-B levels of the antral follicles (26,27) and not a direct measure of follicle quantity or quality.

This study focuses on assigning stages based on rates of FSH change rather than absolute FSH levels, and then in turn, identifies mean ages and amplitudes of FSH associated with intervals with accelerating or decelerating rates of change. If one would transfer the staging vocabulary currently applied to menstrual cycle-defined staging (which is also based on variability), FSH Stage 1 would represent the premenopause and be characterized by minimal change in variability of annual FSH changes. In FSH Stage 2, there was evidence of a major acceleration of FSH rate of change, potentially reflecting an increasing decline in oocyte quality and quantity as more directly assessed by AMH and inhibin-B (28, Sowers MR, Eyvazzadeh AD, McConnell D, Yosef M, Jannausch ML, Harlow S, Randolph JF Jr., Anti-mullerian hormone (AMH) and inhibin-B in defining ovarian aging and the menopause transition, submitted, 2008). In many of the current naming conventions, this might be considered the early perimenopause and according to our observations, this stage would last, on average, 5 years. In FSH Stage 3, there was an acute acceleration in the FSH rate of change which may coincide with the frequent failure to establish a dominant follicle and subsequent corpus luteum and deterioration in the reciprocal relationship of FSH with inhibin-B and inhibin-A, as reviewed by Welt (24). This stage, which might be labeled the late perimenopause, lasted, on average, at least 3 years including approximately 2 years prior to the FMP and 1 year after the FMP. Finally, in FSH Stage 4, commencing 1 year after the FMP, there was a plateau in FSH variability indicating that folliculogenesis was no longer a viable physiological process. It is important to note that these "FSH stages" were developed in relation to a menstrually-defined criterion, the FMP; a one-to-one correspondence between endocrine-based classifications of the menopause transition with a menstrual bleeding-based classification should not be expected, as menstrual bleeding reflects an extensive utero-ovarian biology.

Based on published (6,20) and unpublished data (Sowers MR, Eyvazzadeh AD, McConnell D, Yosef M, Jannausch ML, Harlow S, Randolph JF Jr., Anti-mullerian hormone (AMH) and inhibin-B in defining ovarian aging and the menopause transition, submitted, 2008) describing the declines of AMH and inhibin-B, it is possible to attribute the increasing rate of change in FSH Stage 2 to the loss of restraint by declining inhibin-B levels. However, a definitive explanation of the acute change in FSH in the time period 2 years prior to the FMP has not been satisfactorily articulated, although there are several possible explanations. First, this acute increase in FSH rate of change may be related to a decline in the production of inhibin-A from the corpus luteum. A cross-sectional (29) and longitudinal study (11) suggest that inhibin-A levels eventually decline as women approach the menopausal transition; however, there has been no characterization of an inhibin-A threshold or rate of decline permissive for the ultimate loss of restraint on FSH regulation leading to this observed acute increase in FSH rate of change. A recent study relating inhibin-A levels in women classified into various menopause transition states reported, not unexpectedly, that inhibin-A levels measured in the follicular phase were at or near the limits of assay detection and non-informative (30). Another regulator of FSH is activin A, but recent evidence does not indicate that activin A regulates this acute FSH change (20,30). Alternatively, the loss of regulation could be a function of hypothalamic-pituitary aging rather than the singular loss of control via ovarian-related peptides (31).

Chronological aging represents a population average of the many individual trajectories leading to FMP at variable ages. However, there is additional information to be learned from considering FSH levels in relation to chronological age. For example, these data showed the remarkable stability achieved in FSH levels to age 33 after which time there is a slight increase in the level and variability until age 40 after which time, there is increasing compromise in oocyte quality associated with increasing probability of subfertility and spontaneous abortion. This stability is useful in providing a baseline to appreciate the magnitude of the rate of FSH change at ages 48-52.

The strengths of this report include a large cohort of women representative of a general population group rather than a selected

clinical subsample. There was excellent participation over a 14-year observation period that encompassed the middle and late reproductive periods as well as the menopause transition. The same FSH assay was employed across the time frame with no change in the antibody over time. Further, samples were collected annually and specimens were collected in the early follicular phase of the menstrual cycle. The early follicular phase allows for the comparison of FSH values in a standardized time frame. This data collection protocol, however, precludes evaluation of FSH variation during the later follicular phase which may be more reflective of actual ovulatory events or the luteal phase in which other ovarian peptides, including inhibin-A, may be differentially expressed reflecting endocrine control. Further, this longitudinally acquired data does not include information about hypothalamic-pituitary sensitivity. The population is limited to Caucasian women so findings may not be generalizable to women of other race/ethnic groups. Finally, it would be highly desirable to have these models reproduced in other longitudinally-acquired data.

This report indicates that the natural history of FSH through the late reproductive years and into the menopause transition can be segmented into eight age-specific epochs and four ovarian aging-specific stages by evaluating acceleration and deceleration patterns and grounding these patterns in the underlying biology of oocyte number and folliculogenesis events. These data would indicate that there is a shift from premenopause FSH rate of change stability to early perimenopause stages at age 40 and 42 and that at age 45, there is a major acceleration in the rate of FSH change thought to be the signal that folliculogenesis is increasingly compromised. The reports identifies four FSH stages and the FSH levels and ages that clinicians can use, along with menstrual cycle characteristics, to help interpret the likely status of women with respect to reproductive viability and menopause stage.

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Figure 1. The mean population logFSH (mIU/ml), with 95% upper and lower confidence intervals, in relation to years prior to and following the Final Menstrual Period (FMP) depicting two modeling approaches, a non-parametric stochastic model (black solid line) or a 4-segment piece-wise model (shown with nodes). Vertical reference lines shows the critical nodes associated with time (years) around FMP**.**

Figure 2. Four FSH stages (S1-S4) defined by critical changes in the $_{log}$FSH mIU/ml acceleration and rate of change in relation to the FMP. The acute increasing of $_{log}$FSH mIU/ml (and thus FSH due to the monotonicity of logarithm) occurs between 2 years prior and 1 year after FMP. The major acceleration in logFSH rate of change occurs around 2 years prior to FMP and the major deceleration in logFSH rate of change occurs at 1 year following FMP.

Figure 3. The mean population $_{log}$FSH (mIU/ml), with 95% upper and lower confidence intervals, according to chronological age using two modeling approaches, a non-parametric stochastic model (black solid line) or an 8-segment piece-wise model, with the ages at the nodes where there are critical changes in the line.

Figure 4. Eight age-related FSH segments (A1-A8) defined by critical changes in the $_{log}$FSH mIU/ml acceleration and rate of change. The major acceleration in logFSH rate of change occurs at 45 years of age and the major deceleration in logFSH rate of change occurs at age 55 years.

Table 1. Menopause status, parity, and smoking behavior at the baseline and follow-up visit in 2006/7

		Visit 1992/3	Visit 2006/7
Menopause status			
	Premenopausal	70.6%	23.8%
	Perimenopausal	2.0%	8.4%
	Postmenopausal, natural	--	30.1%
	Postmenopausal, surgical	4.4%	20.2%
	Exogenous hormone use	23.3%	17.5%
Parity			
	Nulliparous	17.4%	14.7%
	Parous, 1~2 live births	50.4%	53.0%
	Parous, > 2 live births	32.3%	32.5%
Smoking			
	Never	57.6%	53.6%
	Ex-smoker	20.1%	31.4%
	Current smoker	22.2%	15.1%

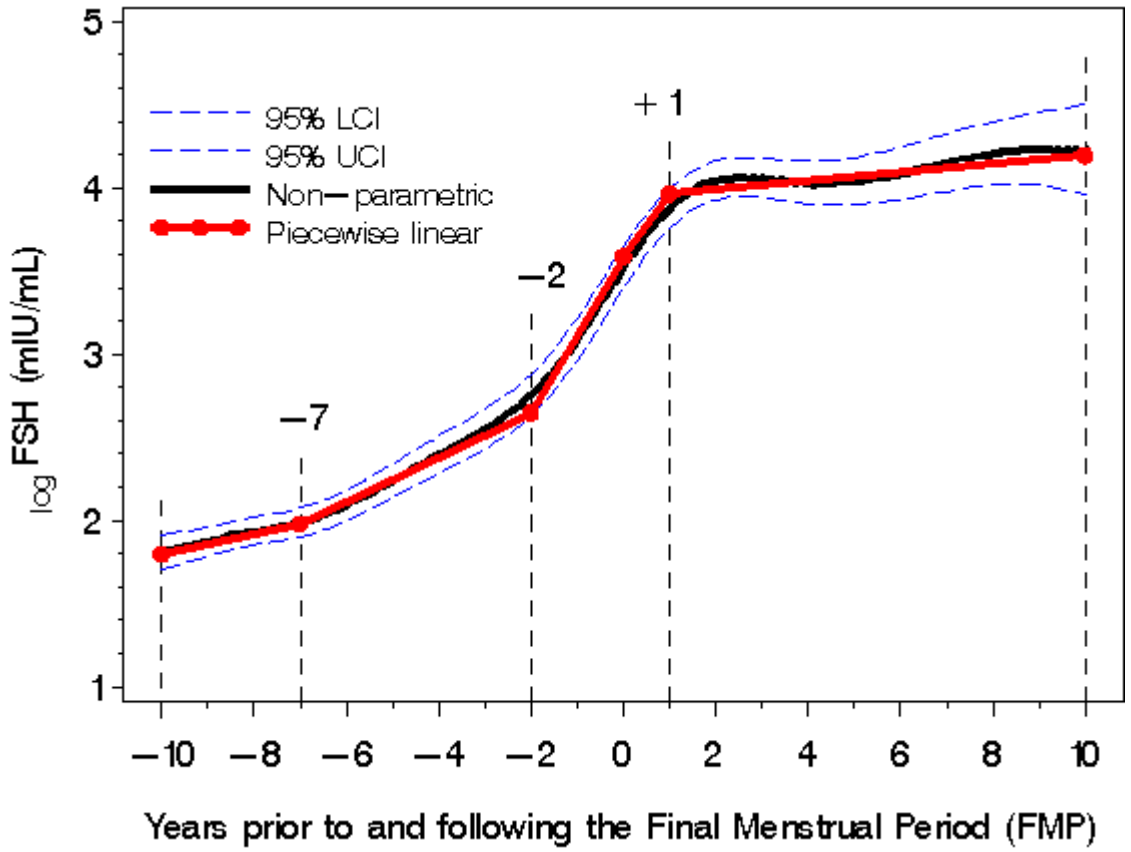
Table 2. Predicted time intervals and age ranges associated with follicle-stimulating hormone (FSH) stages defined by acceleration or deceleration in FSH rate of change related to the final menstrual period (FMP)

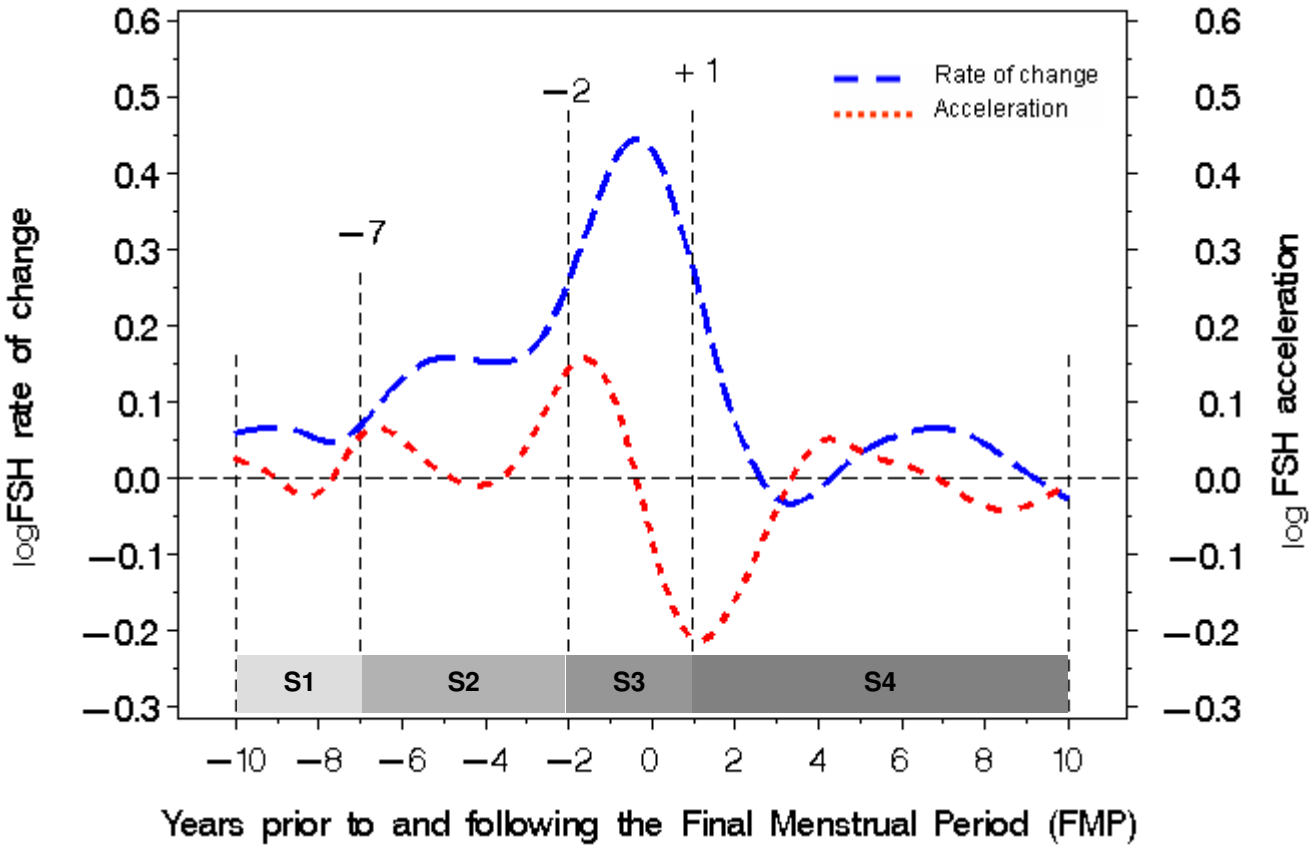
FSH Stages	Time in relation to the FMP	Mean chronologic ages of menopause transition stages
Stage 1	Time prior to -7 years before the FMP	40 to 43.6 years
Stage 2	-7 to -2 years before the FMP	43.6 to 47.6 years
Stage 3	-2 to +1 around the FMP	47.6 to 51.0 years
Stage 4	> 1 year after the FMP	>51.0 years

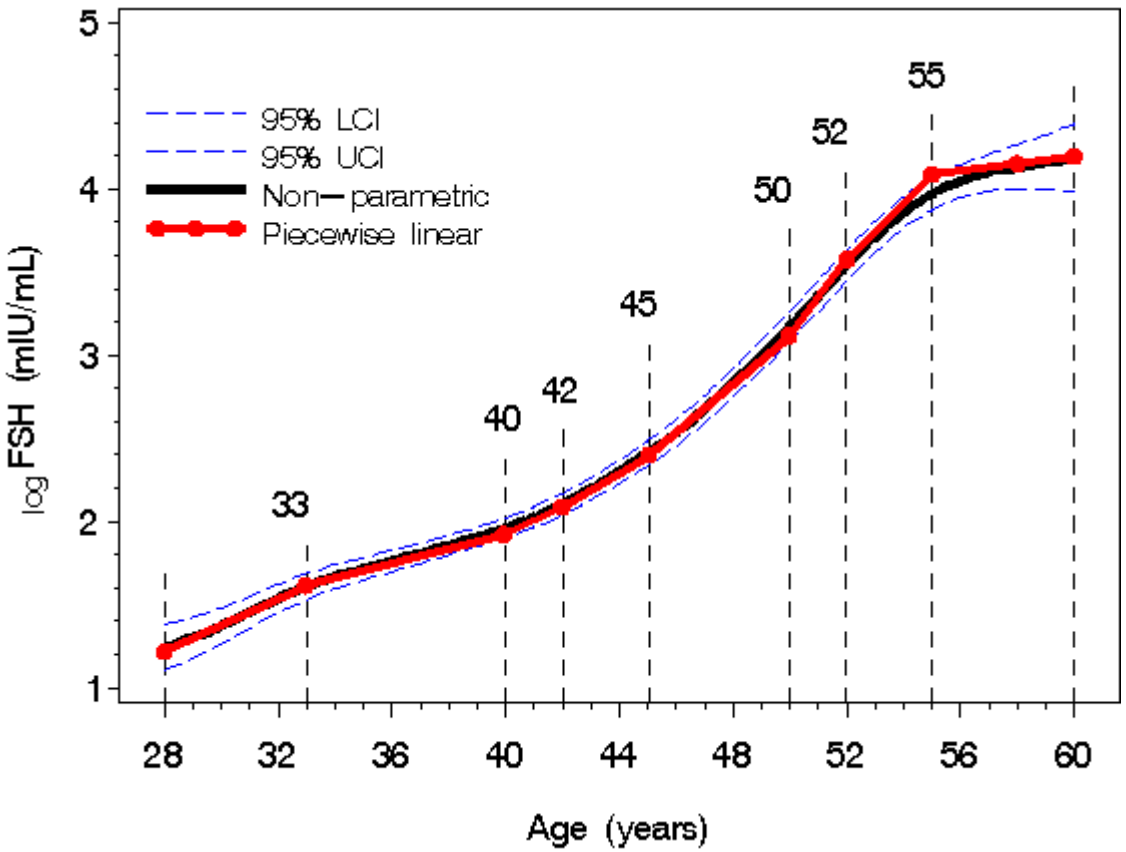
Table 3. Shift in FSH (mIU/ml) levels [mean and standard error (SE) logged and back-transformed], at the critical initial age for each epoch and the percent difference in $_{log}$FSH according to the model fitting approach

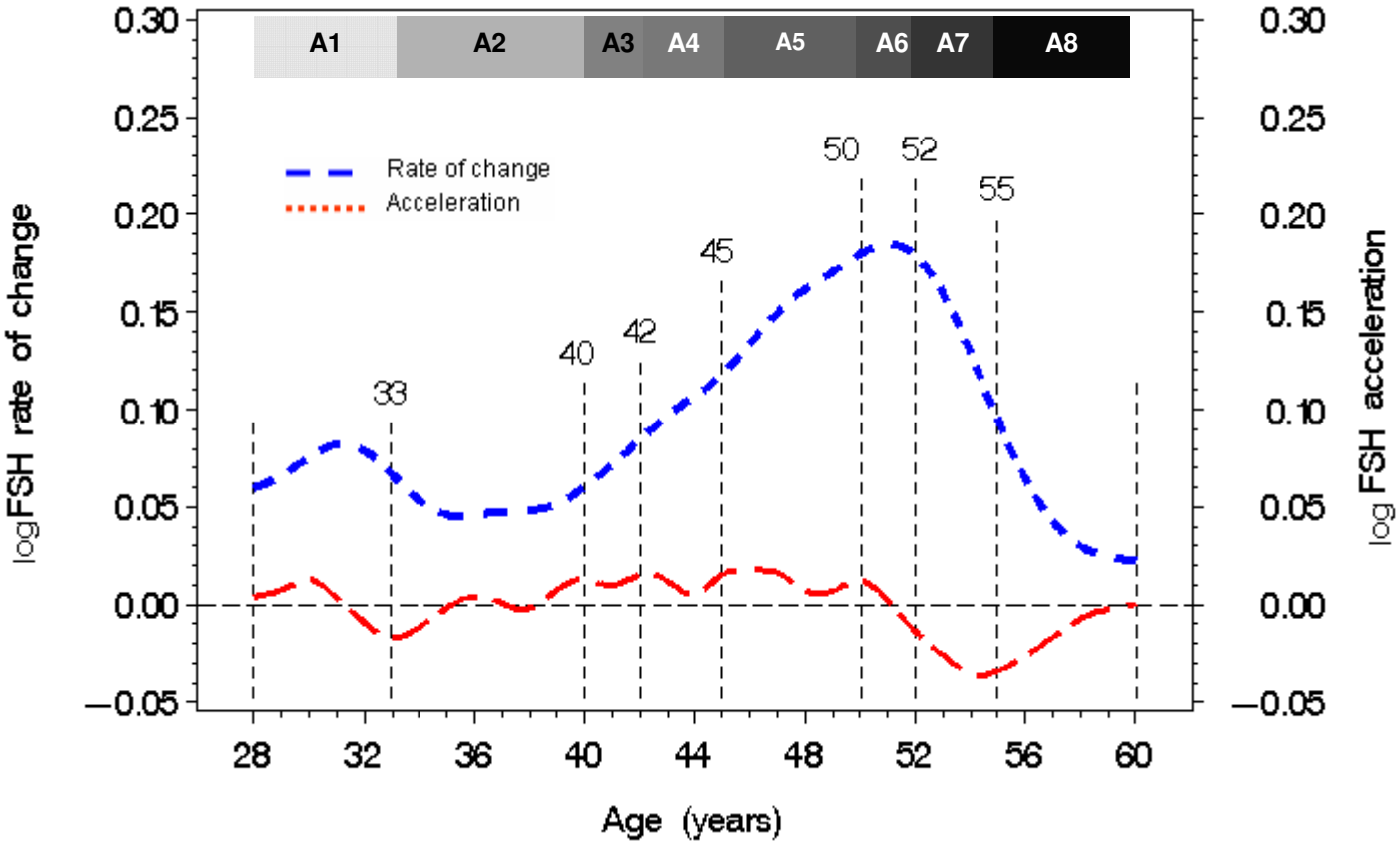
Age	Piece-wise linear mixed model method		Non-parametric stochastic mixed model method		Percent mean $_{log}$FSH differences by two methods[†]
	$_{log}$FSH (SE)	FSH (SE) (back-transformed)	$_{log}$FSH (SE)	FSH (SE) (back-transformed)	
33	1.617 (0.0569)	5.1 (0.29)	1.609 (0.041)	5.0 (0.20)	0.5%
40	1.924 (0.0421)	6.9 (0.29)	1.957 (0.030)	7.1 (0.21)	1.7%
42	2.089 (0.0417)	8.1(0.34)	2.103 (0.032)	8.2 (0.26)	0.7%
45	2.401 (0.0402)	11.1 (0.44)	2.415 (0.038)	11.2 (0.43)	0.6%
50	3.119 (0.0459)	22.7 (1.04)	3.168 (0.046)	23.8 (1.09)	1.5%
52	3.579 (0.0547)	35.9 (1.96)	3.544 (0.047)	34.6 (1.63)	1.0%
55	4.089 (0.0699)	59.8 (4.19)	3.970 (0.049)	53.0 (2.60)	2.9%
60	4.199 (0.1651)	67.6 (11.23)	4.186 (0.106)	65.7 (7.01)	0.3%

[†] The relative differences between two methods were <3% and calculated by $\left|\frac{{}_{\log}FSH^{P} - {}_{\log}FSH^{N}}{{}_{\log}FSH^{N}}\right| \times 100\%$ where the P superscript represents the piecewise linear mixed model method and the N superscript represents the non-parametric stochastic mixed model method.


95% LCI
95% UCI
Non–parametric
Piecewise linear
–7
–2
+1
log FSH (mIU/mL)
5
4
3
2
1
–10
–8
–6
–4
–2
0
2
4
6
8
10
Years prior to and following the Final Menstrual Period (FMP)


0.6
0.5
0.4
0.3
0.2
0.1
0.0
−0.1
−0.2
−0.3
−7
−2
+ 1
Rate of change
Acceleration
S1
S2
S3
S4
−10
−8
−6
−4
−2
0
2
4
6
8
10
logFSH rate of change
log FSH acceleration
Years prior to and following the Final Menstrual Period (FMP)


95% LCI
95% UCI
Non–parametric
Piecewise linear
log FSH (mIU/mL)
Age (years)
1
2
3
4
5
28
32
36
40
44
48
52
56
60
33
40
42
45
50
52
55


A1
A2
A3
A4
A5
A6
A7
A8
Rate of change
Acceleration
33
40
42
45
50
52
55
0.30
0.25
0.20
0.15
0.10
0.05
0.00
−0.05
28
32
36
40
44
48
52
56
60
Age (years)
logFSH rate of change
log FSH acceleration