Original Research

Central, peripheral and resistance arterial reactivity: fluctuates during the phases of the menstrual cycle

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Abstract

The purpose of this study was to document the temporal changes in vascular reactivity occurring simultaneously in central, peripheral and microvascular resistance arteries in the same cohort of women during the normal menstrual cycle. Twenty-three ($n = 23$) women (mean age ($\pm$SD) = 19 $\pm$ 1 y) were tested during four phases of a normal menstrual cycle. Delineation of the four phases occurred as follows: (1) the early follicular phase; (2) the late follicular (LF) phase; (3) the early luteal (EL) phase; and (4) the late luteal phase. Non-invasive measurement of central hemodynamics and peripheral artery pulse wave velocity (PWV) were performed using applanation tonometry. Measurement of peripheral endothelial function was determined by flow-mediated dilation (FMD) testing in the brachial artery and venous occlusion plethysmography in the forearm and calf resistance arteries. Additionally, plasma NO*x* and 17β-estradiol (E) concentrations were measured. Both central (aortic) and peripheral blood pressure (BP) were lowest ($P < 0.05$) during the LF phase and BP reduction was sustained ($P < 0.05$) into the EL phase. The timing and amplitude of the reflected pressure wave were attenuated only during the LF phase ($P < 0.05$). No temporal changes were observed in either central (carotid-femoral) or peripheral PWV (femoral-dorsalis pedis, carotid-radial). Peak forearm and calf blood flow during reactive hyperemia were greatest in LF. Brachial FMD was greatest during the LF phase ($P < 0.05$). Plasma E and NO*x* concentrations were highest during the LF phase ($P < 0.05$). Young premenopausal women experienced an overwhelming pattern of reduced BP and increased systemic vascular reactivity during the LF phase prior to ovulation.

Keywords: menstrual cycle, arterial reactivity, estrogen, nitric oxide

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Introduction

Extensive human data are available regarding the complex endocrine fluctuations during the female menstrual cycle, e.g. increased follicle-stimulating hormone and luteinizing hormone from the anterior pituitary which, in aggregate, stimulate marked changes in the production of estrogens, progesterone and relaxin by the ovarian cells. These changes in the hormonal milieu throughout the menstrual cycle have direct actions on arterial wall physiology, which could play a role in the pathogenesis of hypertension and/or cardiovascular risk in women. However, very limited human data are available regarding the hemodynamic alterations during the discrete phases of the menstrual cycle. A comprehensive assessment of temporal changes in systemic arterial reactivity and wave propagation properties (forward and reflected pressure waves) is the crucial first step for elucidating the mechanism of these alterations. Previous studies have not focused on the entire vascular system including central hemodynamics, central conduit arterial function, muscular resistance arterial function and microvascular arterial function in the same cohort of women.[1–7]

Therefore, the purpose of this study was to document, in detail, the temporal changes occurring simultaneously within the entire vascular system in the same cohort of women both within and between follicular and luteal phases of the normal menstrual cycle. Specifically, we sought to measure temporal trends in central hemodynamics, peripheral muscular artery reactivity and the microvascular resistance artery reactivity. Furthermore, the rapid vasodilating effect of estrogen in endothelial cells is related to its ability to increase nitric oxide (NO) by stimulating endothelial NO synthase activity through activation of the estrogen receptor subgroup alpha.[8–14] Therefore, we also sought to assess, for the first time, the production of NO at four

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time points during the menstrual cycle. We hypothesized that systemic arterial reactivity would mirror the cyclic production of both estrogen and NO during the menstrual cycle.

Methods

Subjects

Apparently healthy, young and eumenorrheic women were recruited. Exclusion criteria included diabetes, reproductive contraceptives, current smoking or hypertension (>135/85 mmHg). None of the subjects were receiving medications for health complications. Each subject tracked the timing of their menstrual phases for one full menstrual cycle prior to the laboratory testing. Each subject was studied at four points during the same menstrual cycle: early follicular (EF) phase two to four days after the onset of menses, late follicular (LF) phase 12 to 14 days after the onset of menses, early luteal (EL) phase 17 to 19 days after the onset of menses and late luteal (LL) phase 25 to 28 days after the onset of menses. Subjects arrived in the laboratory following an 8–12 h overnight fast. Subjects were asked to abstain from caffeine, alcohol and strenuous physical activity for the prior 24 h and to follow the National Institute of Health low-nitrate diet guidelines for the prior 36 h.[15] All vascular testing was performed by the same investigator with patients resting supine in a quiet, temperature-controlled room. The study was approved by the University of Florida Institutional Review Board and written consent was obtained from all subjects.

Blood collection and biochemical analysis

Following a 10-min rest period, venous blood was collected in tubes containing ethylenediaminetetraacetic acid and centrifuged at 3000 rpm for 15 min at 4°C. Plasma was stored at −80°C until analysis at the end of the study. Because NO is a radical gas with a short half-life, measurement of the stable NO metabolites, nitrate and nitrite (NO*x*), was used to estimate NO production. Enzyme-linked immunosorbent assay kits were used to determine plasma concentrations of 17β-estradiol (Calbiotech, Spring Valley, CA, USA) and NO*x* (Cayman Chemical, Ann Arbor, MI, USA).

Central aortic pulse wave analysis: central pressure and hemodynamic measurements

Following a 15-min rest period in a supine position, brachial systolic, diastolic and pulse blood pressure (BP) measurements were performed in triplicate in the left arm using an automated BP cuff (MedTech, Ltd, Vancouver, B.C., Canada). Arterial wave reflection characteristics were obtained using the SphygmoCor system (AtCor Medical, Sydney, Australia). Pressure waveforms were recorded by applanation tonometry of the radial artery using a micromanometer (Millar Instruments, Houston, TX, USA). Optimal recording of the pressure wave was obtained when the waveform was stable for at least 10 beats. A validated generalized mathematical transfer function was used to synthesize a central aortic pressure waveform.[16–20]

The central aortic pressure wave is composed of a forward traveling wave with amplitude ($P_i - P_d$), generated by left ventricular ejection and a reflected wave with amplitude ($P_s - P_i$) that is returning to the ascending aorta from the periphery.[21] The contribution of the reflected wave to ascending aortic pulse pressure can be estimated by the aortic augmentation index (AI_a). AI_a is defined as reflected wave amplitude divided by pulse pressure and expressed as a percentage ($AI_a = [P_s - P_i]/[P_s - P_d] \times 100$).[21,22] P_s is the peak systolic pressure, P_i is the beginning upstroke of the reflected pressure wave and P_d is the minimum diastolic pressure. Due to heart rate (HR) effects on pressure wave variability, AI_a was normalized to HR of 75 bpm (AI_a at 75 bpm). The round-trip travel time (Δt_p) of the forward traveling wave from the ascending aorta to the major reflection site and back is measured from the foot of the forward traveling pressure wave to P_i. Δt_p is directly related to the distance to the reflecting site.[21] The augmented pressure produced by the reflected wave ($P_s - P_i$) and its duration (ejection duration–Δt_p) decreases aortic blood flow.[21] Wasted left ventricular energy (LVEw) is the quantification of excess energy expended by the left ventricle without improving cardiac output ($LVEw = [(\pi/4) \cdot (P_s - P_i) \cdot (\text{ejection duration} - \Delta t_p) \cdot 1.333]$) and was expressed as dynes/cm^2/s.[23]

Pulse wave velocity: central and peripheral arterial stiffness measurements

Pressure waveforms were gated with simultaneous electrocardiographs and were used to calculate the pulse wave velocity (PWV) between two sites. Tonometry transit distances from the supra-sternal notch to the radial (SSN-R), femoral (SSN-F) and carotid (SSN-C) and from the femoral to the dorsalis pedis (F-DP) recording site were measured with a tape measure.[24] Foot-to-foot PWV was calculated by determining the delay between the appearance of the pressure waveform foot between two sites (Δt), using a SphygmorCor Pulse Wave Velocity Vx system and SCOR-2000 Version 6.31 software (Atcor, Sydney, Australia).[24] The distance between recording sites was adjusted for parallel transmission in the aorta and carotid by subtracting SSN-C from SSN-R and SSN-F. The corrected distances were divided by the respective foot-to-foot transmission delays (carotid-radial, carotid-femoral [C-F]) to give PWV. Central PWV was evaluated using the C-F data and peripheral PWV using the femoral-dorsalis pedis (F-DP) and carotid-radial (C-R) data. PWV was used as an indirect measure of regional arterial stiffness.

Peripheral flow-mediated dilation: conduit artery shear-induced or endothelial-dependent reactivity measurement

Brachial artery flow-mediated dilation (FMD) was assessed in the right arm using a high-resolution ultrasound machine (ATL HDI 3000; Advanced Technologies Laboratories, Bothell, WA, USA) equipped with a 10.5-MHz transducer as recently described in a Technique Report by the International Brachial Artery Reactivity Task Force.[25] Briefly, resting baseline end diastolic brachial diameters and

blood velocity were obtained with the transducer placed 3–5 cm above the anticubital fossa. Reactive hyperemia was produced by inflating a BP cuff placed on the upper forearm for 5 min at 200 mmHg followed by a rapid deflation. The brachial artery was imaged and recorded for three minutes following cuff deflation. Ultrasound images were recorded on a super VHS videocassette for offline electronic image analysis using Image Pro Software (Image Pro, Data Translation, Inc, Marlboro, MA, USA). Brachial artery diameters were determined during end-diastole (gated with electrocardiogram R wave) by measuring the distance between the near and far wall of the intima. Peak brachial FMD was expressed as a percentage increase from baseline (FMD%). Because peak FMD% is influenced by baseline diameter, absolute changes (Δmm) in diameter were also determined.[26] Moreover, all measurements were normalized to the mean shear rate calculated from the first 10 s following cuff dilation.

Forearm and calf blood flow: microvascular resistance artery shear-induced or endothelial-dependent reactivity measurement

Venous occlusion plethysmography (EC-6, D.E. Hokanson, Inc, Bellevue, WA, USA) using calibrated mercury strain gauges was used to determine forearm blood flow (FBF) and calf blood flow (CBF).[27] Briefly, the left forearm or left calf was positioned above heart level and a strain gauge was placed around the widest part of the non-dominant forearm or calf. A cuff placed on the upper arm or thigh was inflated to 50 mmHg for seven seconds to prevent venous outflow.[27] The venous collecting pressure was set at 50 mmHg based on evidence demonstrating that this collecting pressure yields the most accurate measurements of flow during reactive hyperemia.[28] During measurement of FBF and CBF, circulation to the hand or foot was arrested by inflation of a cuff placed around the wrist and ankle and inflated to suprasystolic levels (220 mmHg). The plethysmograph output signal was transmitted to a calibrated software program (NIVP3, D.E. Hokanson, Inc) on a laptop PC computer and the slope was expressed as milliliters per minute per 100 mL of tissue (mL/min per 100 mL tissue). Absolute blood flow was determined by the rate of change of limb circumference (e.g. slope) during the seven-second venous occlusion, which correlates highly to arterial blood inflow into the limb.[29,30] Flow was measured four times each minute for two minutes and averaged to represent resting FBF and CBF. In an attempt to measure the endothelial-dependent reactivity of the microcirculation, we occluded blood flow for 5 min using a cuff inflated to 200 mmHg and positioned proximal to the anticubital fossa of the arm, or proximal to the knee.[31,32] Peak blood flow after 5 min of ischemia was recorded as the highest FBF or CBF observed immediately following cuff release. Total blood flow for 3 min was defined as the area under the blood flow–time curve after baseline flow was subtracted.[32]

Statistical analysis

The descriptive data are expressed as means ± SD. Comparisons across the four phases of the menstrual cycle were made using one-way analysis of variance with repeated measures. *Post hoc* analysis was performed using appropriate least significant difference tests. The alpha level for statistical significance was set at 0.05 and data were analyzed using SPSS Version 17.0 (SPSS, Chicago, IL, USA).

Results

Subject descriptive characteristics

A total of twenty-three ($n = 23$) women (mean [±SD] age = 19.7 ± 1.3 y; height = 165 ± 6 cm; body mass = 61.8 ± 15.4 kg; body mass index (BMI) = 22.4 ± 4.0 kg/m^2) completed laboratory vascular testing during the four menstrual phases. Body mass and BMI did not differ across the four measurement time points.

Blood pressure and pulse wave analysis

We observed no significant change in HR across the four phases of the menstrual cycle. Peripheral and central BP data are presented in Figure 1. Peripheral systolic and diastolic BP, and aortic systolic and diastolic BP were all lower ($P < 0.05$) during the LF phase when compared with the

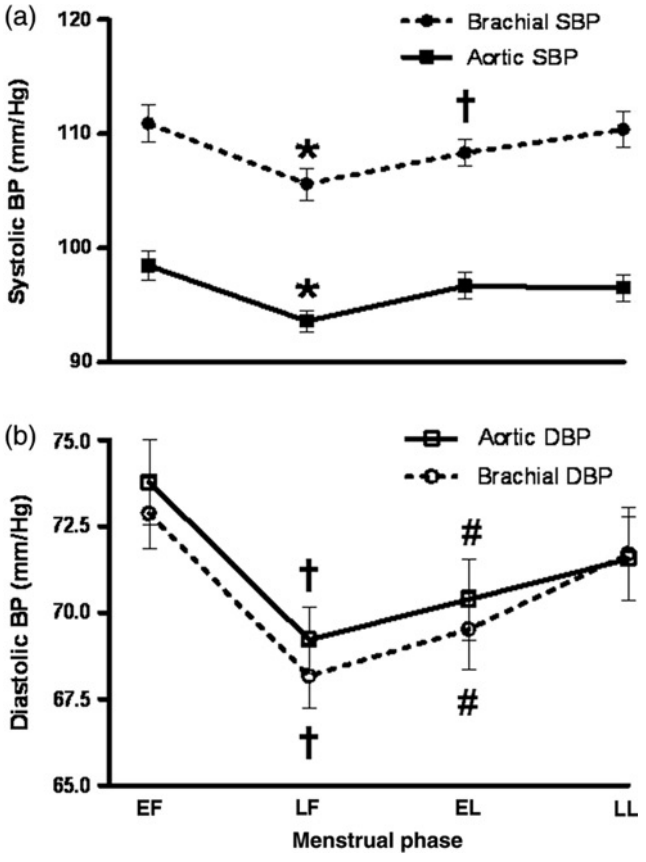

Figure 1 (Top panel) Temporal pattern of brachial and aortic systolic blood pressure during the early follicular (EF), late follicular (LF), early luteal (EL) and late luteal (LL) phases of the menstrual cycles. (Bottom panel) Temporal pattern of brachial and aortic diastolic blood pressure during the EF, LF, EL and LL phases of the menstrual cycle. $^{*}P < 0.05$ versus EF, EL and LL; $^{\dagger}P < 0.05$ versus EF and LL; $^{\#}P < 0.05$ versus EF. SBP, systolic blood pressure; DBP, diastolic blood pressure

EF and LL phases. Peripheral systolic and diastolic BP, and aortic diastolic BP were all lower ($P < 0.05$) during the EL phase when compared with the EF phase (Figure 1).

Pulse wave analysis (PWA) data are presented in Figure 2. AI_a (6.9 ± 8.7 versus 14.0 ± 8.7, 11.9 ± 7.5 and $12.5 \pm 9.2\%$) and AI_a at 75 bpm were lower ($P < 0.05$) during the LF phase when compared with the EF, EL and LL phases, respectively. Round trip travel time of the reflected wave (Δt_p) was increased ($P < 0.05$) during LF and EL when compared with EF and LL. Augmented BP (1.5 ± 2.1 versus 3.4 ± 2.2, 2.8 ± 1.9 and 3.0 ± 2.9 mmHg) and wasted left ventricular energy were lower ($P < 0.05$) in LF when compared with EF, EL and LL, respectively. Additionally, Δt_p remained increased ($P < 0.05$) and wasted left ventricular energy remained lower ($P < 0.05$) during the EL phase, when compared with EF.

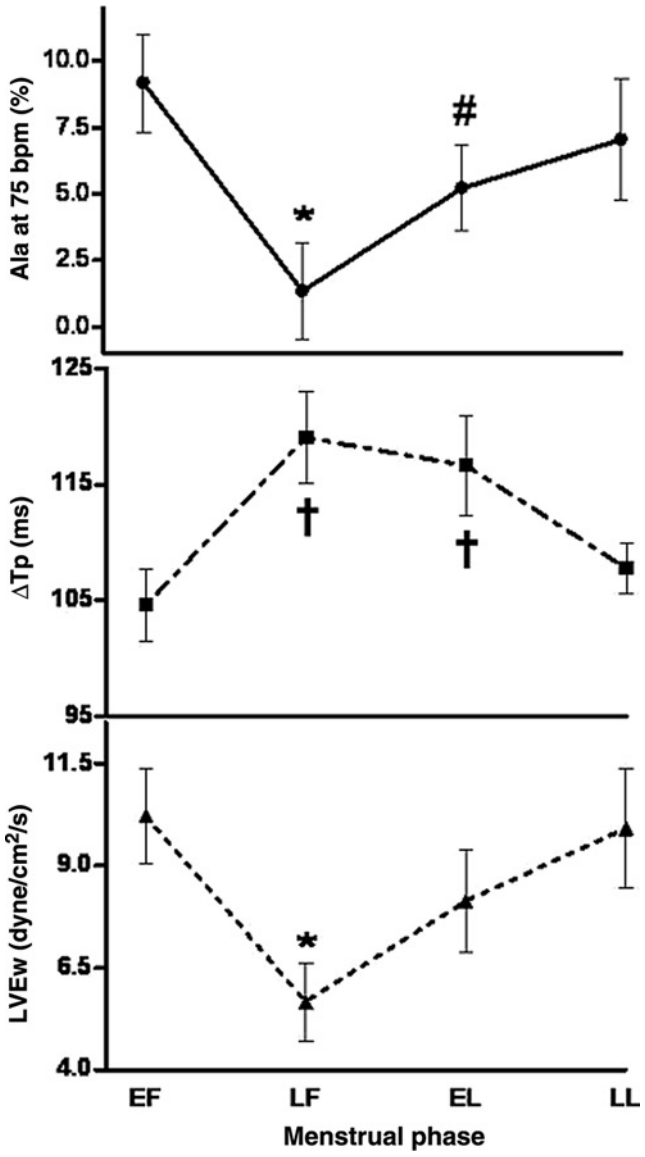

Figure 2 Temporal pattern of the percent change in round trip travel time of the aortic pressure wave (ΔTp; ms), wasted left ventricular energy (LVEw; dyne/cm^2/s) and aortic augmentation index normalized for a heart rate of 75 bpm (Aix at 75; %) during the early follicular (EF), late follicular (LF), early luteal (EL) and late luteal (LL) phases of the menstrual cycle. $^*P < 0.05$ versus EF, EL and LL; $^\dagger P < 0.05$ versus EF and LL; $^\#P < 0.05$ versus EF

Central and peripheral arterial stiffness

Central and peripheral pulse wave velocity (PWV) results are presented in Table 1. No significant changes were observed in either central (carotid-femoral, carotid-radial) or peripheral (femoral-dorsalis pedis) PWV across the four phases of the menstrual cycle.

Forearm and calf resistance arterial blood flow

Forearm and CBF data are presented in Table 2. Resting FBF and CBF values did not differ across the menstrual cycle. However, peak FBF and CBF during reactive hyperemia were greater ($P < 0.05$) in LF when compared with EF, EL and LL. Additionally, total FBF and CBF area under the curve at 3 min was greater ($P < 0.05$) in LF when compared with EF, EL and LL.

Brachial artery endothelial function

Brachial artery FMD results are shown in Figure 3. Absolute dilation (mm), brachial FMD (%) and FMD normalized during the first 10 s following cuff release (s^{-1}) were all greater ($P < 0.05$) during the LF phase, when compared with the EF, EL or LL phases. There were no significant changes in baseline brachial diameter (mm).

Table 1 Central and peripheral pulse wave velocity

	EF	LF	EL	LL
CFPWV (m/s)	6.5 ± 0.6	6.1 ± 0.6	6.4 ± 0.5	6.4 ± 0.4
CRPWV (m/s)	9.5 ± 1.1	8.9 ± 1.1	9.2 ± 1.2	9.3 ± 1.2
FDPWV (m/s)	8.7 ± 1.5	8.2 ± 1.4	8.5 ± 1.3	8.6 ± 1.2

Values are mean ± SD
EF, early follicular: LF, late follicular; EL, early luteal; LL, late luteal; CFPWV, carotid-femoral pulse wave velocity; CRPWV, carotid-radial pulse wave velocity; FDPWV, femoral-distal pulse wave velocity

Table 2 Forearm and calf resistance arterial blood flow

	EF	LF	EL	LL
Peak FBF (mL/min/100 mL tissue)	21.5 ± 04.3	$25.8 \pm 05.9^*$	22.3 ± 06.9	21.5 ± 5.7
Peak CBF (mL/min/100 mL tissue)	26.3 ± 4.2	$29.0 \pm 5.4^*$	26.9 ± 6.6	26.2 ± 4.2
Total FBF (AUC_{3min})	6.1 ± 1.9	$7.6 \pm 2.9^*$	6.4 ± 2.0	6.1 ± 1.8
Total CBF (AUC_{3min})	7.2 ± 1.3	$8.9 \pm 2.5^*$	7.6 ± 1.9	7.5 ± 1.7

EF, early follicular: LF, late follicular; EL, early luteal; LL, late luteal; FBF, forearm blood flow; CBF, calf blood flow; AUC, area under the curve
Values are mean ± SD; peak FBF = peak forearm blood flow; peak CBF = peak calf blood flow; total FBF = total forearm flow area under the curve at three minutes; total CBF = total forearm flow area under the curve at three minutes
$^*P < 0.05$ versus EF, EL and LL

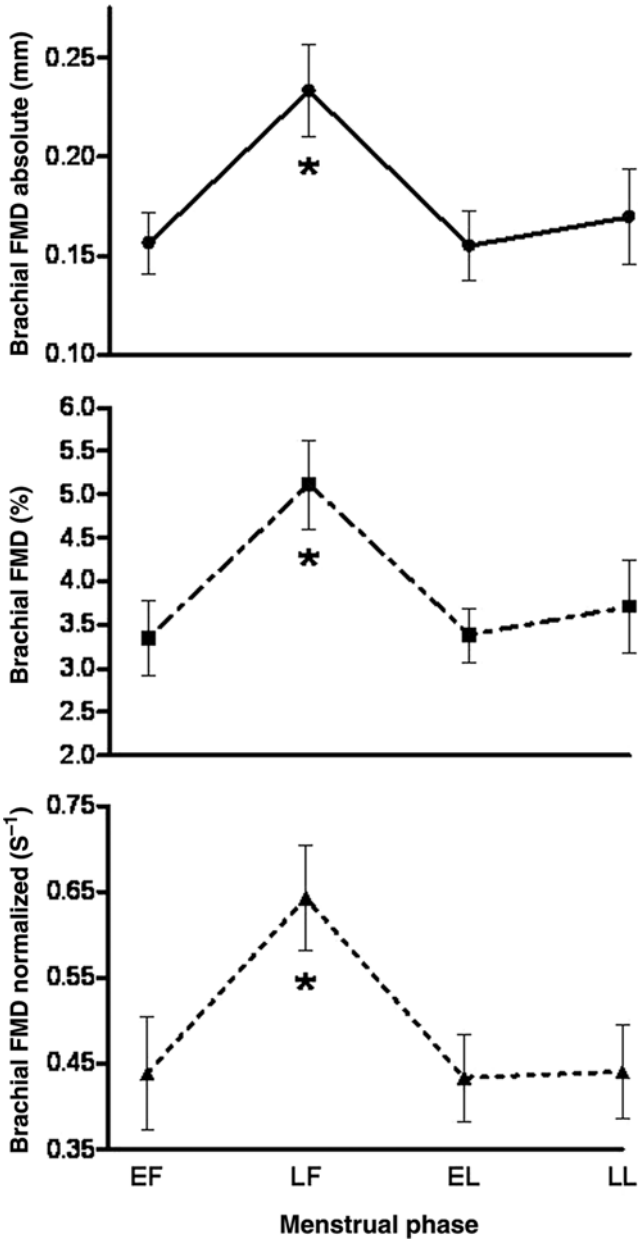

Figure 3 Temporal pattern of absolute dilation (mm) (top panel), percent dilation (%) (middle panel) and dilation normalized during the first 10 s following cuff release (bottom panel) during flow-mediated dilation (FMD) testing in the brachial artery during the early follicular (EF), late follicular (LF), early luteal (EL) and late luteal (LL) phases of the menstrual cycle. $^{*}P < 0.05$ versus EF, EL and LL

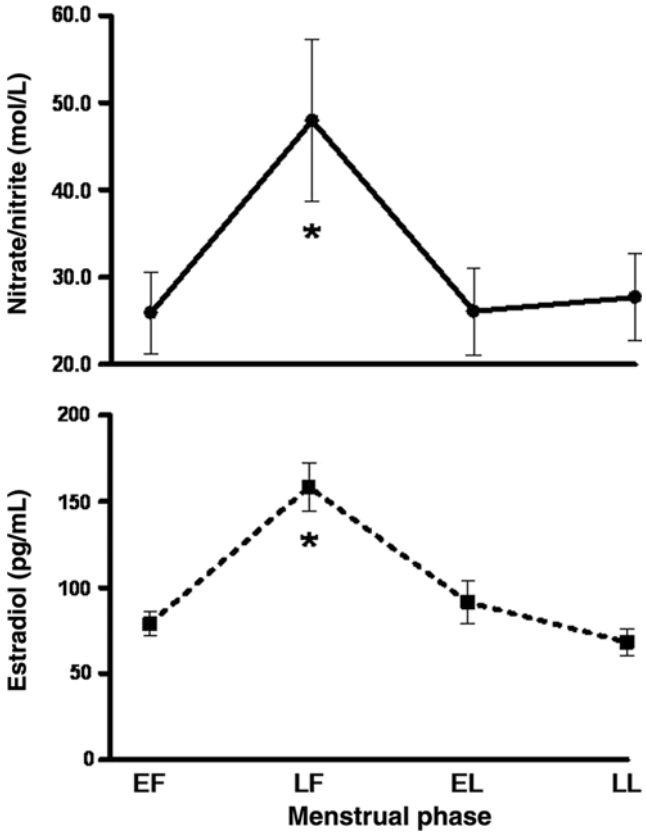

Figure 4 Temporal pattern of plasma concentrations of nitrite/nitrate (NO*x*) (top panel) and 17β-estradial (bottom panel) during the early follicular (EF), late follicular (LF), early luteal (EL) and late luteal (LL) phases of the menstrual cycle. $^{*}P < 0.05$ versus EF, EL and LL

Nitric oxide and estrogen

NO*x* and estradiol data are presented in Figure 4. Plasma NO*x* concentrations were greater ($P < 0.05$) during the LF phase when compared with the EF, EL or LL phases. Plasma estradiol concentrations were greater ($P < 0.05$) during the LF phase when compared with the EF, EL or LL phases.

Discussion

The first principal finding of the present study was that the entire vascular system is affected by the four phases of the menstrual cycle. Central (aortic) and systemic BPs were decreased toward the end of the follicular (LF) phase and into the EL phase when compared with the other phases. Peripheral vascular reactivity of muscular arteries and the microvasculature was increased toward the end of the follicular phase (LF) when compared with the other three phases. The second principal finding of the present study was that temporal changes in BP and peripheral vascular reactivity perfectly mirrored the cyclic production of both estrogen and NO during the menstrual cycle (Figures 1–4). To our knowledge, this was the first study that has measured NO throughout the four phases of the menstrual cycle in the same cohort of women.

Our data suggest that NO production may be a primary mechanism driving temporal fluctuations in arterial reactivity in premenopausal women. Indeed, NO concentrations increased by approximately 85% between the early (EF) and late (LF) follicular phases and returned toward baseline levels during the luteal phases (EL, LL) (Figure 4). Both endothelial and vascular smooth muscle cells contain estrogen receptors.[8,9,33–35] Binding of estrogen to these receptors activates pathways that subsequently increase NO bio-availability.[8,10,13,14] It seems reasonable to hypothesize that the increased production of estrogen during the LF phase results in a greater opportunity for estrogen receptor binding, thereby causing an increase in NO bioavailability. NO is an important modulator of vascular tone[36] and

vascular reactivity[25,37] and this estrogen-mediated increase in NO is a possible mechanism responsible for increased vascular reactivity during the LF phase. However, the present study was not designed to establish a direct causal link between estrogen concentrations and NO production and this relationship requires further study. It is noteworthy, however, that the relationship between plasma concentrations of NO*x* and peak brachial FMD responses in the present cohort was similar to what we have previously reported ($r = 0.53$; $P = 0.001$) in young men and women.[38]

Endothelial-mediated brachial artery FMD increased by 53% during the LF phase but returned rapidly toward baseline values during the EL phases (Figure 3). Consistent with the present study, Williams *et al.*[1] and Hashimoto *et al.*[2] reported significant increases in brachial FMD between the EF and LF phases. However, those studies also reported a second significant increase in brachial FMD during the luteal phase.[1,2] The explanation for their biphasic brachial FMD results is unclear. We speculate, however, that the luteal brachial FMD data in those studies were collected too near the LF phase. Indeed, both studies reported estrogen concentrations at the timing of the luteal brachial FMD measurements that were not significantly decreased from values recorded at the LF phase.[1,2]

Endothelial function in resistance arteries also demonstrated a temporal pattern of fluctuation during the four phases of the menstrual cycle. Peak FBF and peak CBF increased by 20% and 12%, respectively, between the EF phase and the LF phase. Both peak FBF and peak CBF returned rapidly to baseline values during the EL phase. To the best of our knowledge, this is the first study to assess microcirculation resistance artery function at multiple time points throughout the menstrual cycle.

Lastly, central hemodynamics also showed a temporal pattern of fluctuation during the four phases of the menstrual cycle. Both the amplitude and timing of the aortic reflected pressure wave were significantly decreased during the LF phase, when compared with EF and LL phases (Figure 2). Aortic augmentation (AI_a at HR = 75) decreased by 85% and round trip travel time of the reflected wave (Δt_p) increased by 14% between the EF and LF phases (Figure 2). The observed reductions in reflected pressure led to a 33% decrease in wasted left ventricular energy during the LF phase (Figure 2). These BP and hemodynamic changes persist for approximately 10 to 14 days (LF through EF phases) and may confer protection against cardiovascular risk. Moreover, these findings may have implications for women's reproductive health. The observed improvement in central and peripheral hemodynamics during the LF phase may provide a better vascular environment for endometrial receptivity and egg implantation.[39] Indeed, the expansion of the endometrial vascular bed observed between the LF and EL phases may, in turn, be one mechanism responsible for cyclic changes in central hemodynamics. Increased endometrial vascularity produces a blood buffering effect that would shift the reflecting sites distally and change the timing and amplitude of aortic reflected pressure waves.[40] To date, only one other study has examined central reflected wave characteristics in premenopausal women.[5] Ounis-Skali *et al.*[5] failed to observe significant differences in AI_a. However, they collected data on only two occasions: once during the EF phase and once during the EL phase. It is important to note that in the present study we also observed no differences in AI_a between the EF and EL phases. However, when central hemodynamic data were analyzed from the four discrete phases of the menstrual cycle, we observed robust decreases in aortic augmented pressure during the LF phase that differed significantly from the EF, EL and LL phases.

The present study revealed no temporal changes in central or peripheral arterial stiffness, when measured by PWV (Table 1). This is in agreement with all previous studies that have measured PWV throughout the menstrual cycle.[1,4,5] However, we did observe a significant temporal effect on systemic BP across the four phases of the menstrual cycle, consistent with results from 24-h ambulatory monitoring.[7] Peripheral systolic and diastolic BP, and aortic systolic and diastolic BP all decreased by approximately 4 mmHg during the LF phase and were significantly lower than during the EF, EL and LL phases (Figure 1). Our data clearly demonstrate the importance of standardizing the timing of vascular testing in premenopausal women in the research laboratory. Perhaps more importantly, our BP data may have clinical relevance. Although the BP reductions observed during the LF and EL phases in our eumenorrheic subjects seem modest (~4 mmHg reduction for the duration of 10 to 14 days), a systolic BP reduction of only 3 mmHg in average populations has been estimated to reduce cardiac morbidity by 5–9%, stroke by 8–14%, and all cause mortality by 4%.[41] In individual study subjects, the BP reduction during the LF and EL phases was greater and it is estimated that a 10 mmHg reduction in systolic BP or a 5 mmHg reduction in diastolic BP could reduce overall cardiovascular risk by approximately 50%.[41] In aggregate, our data suggest that menstrual cycle phase should be a consideration when measuring BP in the clinical setting.

Cardiovascular disease in premenopausal women is conspicuously less than in men of comparable age.[42] Cyclical elevations in estrogen are thought to be one mechanism responsible for this phenomenon. Our data further suggest that LF spikes in NO production may confer some degree of cardiovascular 'protection' in women. In addition to being an important vasoactive agent, NO inhibits leukocyte adhesion, platelet aggregation, expression of adhesion molecules, endothelin-1, smooth muscle cell proliferation and endothelial inflammation. The monthly LF surge in estrogen and NO throughout the menstrual years, and the consequent reduction in BP for 10 to 14 days, may reduce risk of cardiovascular disease.

In conclusion, BP, central hemodynamics and reactivity of peripheral and microvascular arteries improve significantly during the LF phase prior to ovulation and returns to baseline in the LL phase. The mechanism responsible for improved arterial function in the LF phase appears to be, at least in part, estrogen-mediated increases in NO bioavailability. These data highlight the importance of standardizing the timing of vascular testing in premenopausal women in the research laboratory and the importance of

considering the menstrual cycle phase when measuring BP in the clinical setting.

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