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Markers of vitamin D metabolism and premenstrual symptoms in healthy women with regular cycles

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STUDY QUESTION: Are insufficient 25-hydroxyvitamin D (25(OH)D) concentrations, and other markers of vitamin D metabolism, associated with premenstrual symptoms in healthy women with regular menstrual cycles?

SUMMARY ANSWER: 25(OH)D insufficiency was associated with specific physical premenstrual symptoms, while no associations were observed with psychological symptoms or with other markers of vitamin D metabolism.

WHAT IS KNOWN ALREADY: Prior studies evaluating vitamin D and premenstrual symptoms have yielded mixed results, and it is unknown whether 25(OH)D insufficiency and other markers of vitamin D metabolism are associated with premenstrual symptoms.

STUDY DESIGN, SIZE, DURATION: We used two cohorts of women with regular menstrual cycles; 1191 women aged 18–40 years in EAGeR (cross-sectional analysis of a prospective cohort within a randomized trial) and 76 women aged 18–44 years in BioCycle (prospective cohort). In EAGeR, premenstrual symptoms over the previous year were assessed at baseline, whereas in BioCycle, symptoms were assessed prospectively at multiple points over two menstrual cycles with symptoms queried over the previous week. In both cohorts, symptomatology was assessed via questionnaire regarding presence and severity of 14 physical and psychological symptoms the week before and after menses. Both studies measured 25(OH)D in serum. We also evaluated the association of additional markers of vitamin D metabolism and calcium homeostasis, including intact parathyroid hormone (iPTH), calcium (Ca), fibroblast growth factor 23 (FGF23), and 1,25 dihydroxyvitamin D ($1,25(OH)_2D$) with premenstrual symptoms in the BioCycle cohort.

PARTICIPANTS/MATERIALS, SETTING, METHODS: One cohort of women actively seeking pregnancy (Effects of Aspirin in Gestation and Reproduction (EAGeR)) and one cohort not seeking pregnancy (BioCycle) were evaluated. Log-binomial regression was used to estimate risk ratios (RR) and 95% CIs for associations between insufficient 25(OH)D (<30 ng/ml) and individual premenstrual symptoms, adjusting for age, BMI, race, smoking, income, physical activity, and season of blood draw.

MAIN RESULTS AND THE ROLE OF CHANCE: 25(OH)D insufficiency was associated with increased risk of breast fullness/tenderness (EAGeR RR 1.27, 95% CI 1.03, 1.55; BioCycle RR 1.37, 95% CI 0.56, 3.32) and generalized aches and pains (EAGeR RR 1.33, 95% CI 1.01, 1.78; BioCycle 1.36, 95% CI 0.41, 4.45), though results were imprecise in the BioCycle study. No associations were observed between insufficient 25(OH)D and psychological symptoms in either cohort. In BioCycle, iPTH, Ca, FGF23, and $1,25(OH)_2D$ were not associated with any premenstrual symptoms.

LIMITATIONS, REASONS FOR CAUTION: Results from the EAGeR study were limited by the study design, which assessed both 25(OH)D at baseline and individual premenstrual symptoms over the past year at the baseline. As such, reverse causality is a potential

concern. Though premenstrual symptoms were assessed prospectively in the BioCycle cohort, the power was limited due to small sample size. However, results were fairly consistent across both studies.

WIDER IMPLICATIONS OF THE FINDINGS: Serum 25(OH)D may be associated with risk and severity of specific physical premenstrual symptoms.

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Introduction

Premenstrual syndrome (PMS) affects approximately 50% of reproductive age women globally (Direkvand-Moghadam *et al.*, 2014) and is characterized by physical and psychological symptoms such as breast tenderness, headaches, bloating, irritability, anxiety, depression, and mood swings (Halbreich *et al.*, 2003; Sternfeld *et al.*, 2002). These symptoms range in severity and can negatively affect normal day-to-day life, including productivity and interpersonal relationships (Borenstein *et al.*, 2003).

Non-pharmaceutical therapies for premenstrual symptoms, particularly related to dietary intake, are of interest as they offer potential low-cost strategies to improve symptomology. Vitamin D has been proposed as one such treatment as it has been shown to improve dysmenorrhea (Lasco *et al.*, 2012), perhaps via a reduction in proinflammatory cytokines and prostaglandins which are essential for menstrual cycle function (Krishnan and Feldman, 2011). Vitamin D may also be related through calcium homeostasis, as low calcium intake is associated with higher risk of PMS (Bertone-Johnson *et al.*, 2005). Additionally, low serum concentrations of 25-hydroxyvitamin D (25(OH)D) have been associated with higher rates of mood disorders, specifically depression (Hoang *et al.*, 2011), a psychological factor associated with PMS (Hofmeister and Bodden, 2016). Calcium intake and calcium homeostasis have also been linked to PMS (Thys-Jacobs, 2000), suggesting the importance of also considering other markers of vitamin D metabolism. Calciotropic hormones, including intact parathyroid hormone (iPTH), calcium, fibroblast growth factor 23 (FGF23), and 1,25 dihydroxyvitamin D (1,25(OH) $_2$D) may also play a role and offer important insight, as hypocalcemia is known to cause symptoms that are similar to those that occur in PMS, such as muscle cramps, irritability, anxiety, and fatigue (Schafer, 2016). Thus, 25(OH)D and other markers of vitamin D metabolism may be associated with premenstrual symptoms.

Prior studies evaluating vitamin D and PMS and premenstrual symptoms have yielded mixed results (Bertone-Johnson *et al.*, 2010). Among women in the Nurses' Health Study II (NHS2), Bertone-Johnson *et al.* (2010) found that higher dietary intakes of vitamin D were associated with lower risk of PMS, though low plasma 25(OH)D concentrations were not associated with incident PMS among women over 30 years of age in the same population (Bertone-Johnson *et al.*, 2014), with similar results observed in a cohort of college-age women (Bertone-Johnson *et al.*, 2010). However, in the NHS2 cohort, increasing plasma 25(OH)D concentrations were associated with lower risk of specific premenstrual symptoms, including breast tenderness, fatigue, diarrhea/constipation, and depression, suggesting a potential role for 25(OH)D in symptom management (Bertone-Johnson *et al.*, 2014). Importantly, these prior studies examined associations of vitamin D and the initial development of PMS; it remains unclear whether 25(OH)D status may influence cycle-specific risk of individual premenstrual symptoms. Furthermore, the role of other markers of vitamin D metabolism and risk of premenstrual symptoms is largely unknown.

Thus, we assessed associations between multiple markers of vitamin D metabolism, including 25(OH)D, iPTH, calcium, FGF23, and 1,25(OH) $_2$D with physical and psychological premenstrual symptoms, among two cohorts of women aged 18–44 years with regular menstrual cycles.

Materials and methods

Study designs and populations

We conducted our analysis among two cohorts of women between the ages of 18 and 44 years old. The first cohort included 1191 women enrolled in the Effects of Aspirin in Gestation and Reproduction (EAGeR) trial, a multicenter, block-randomized, double-blind, placebo-controlled trial to assess the effects of low-dose aspirin (81 mg) on live birth and pregnancy loss in four US medical centers from 2006 to 2012 (Schisterman *et al.*, 2013). Participants were recruited via provider/clinic-based recruitment involving brochures in the clinic, waiting rooms, and restrooms of hospitals and clinics, private practices, and community health centers throughout the catchment areas (Lesher *et al.*, 2015). Initially, 5485 women contacted the study and were assessed for eligibility; of these, 4257 (78%) were excluded (4088 not eligible, 60 discontinued after baseline assessment, 109 tested positive for pregnancy before randomization), and 1228 (22%) women were randomized and enrolled in the trial (Schisterman *et al.*, 2014). Of these, 1191 (97%) had 25(OH)D measured at baseline. The present analysis involves a cross-sectional baseline assessment of 25(OH)D and premenstrual symptoms from this cohort. Women

who were trying to conceive, were 18–40 years old, with self-reported regular menstrual cycles of 21–42 days in length, no known history of infertility, 1 or 2 documented prior pregnancy losses, and had up to 2 prior live births were eligible. Exclusion criteria included but were not limited to (a) bleeding disorders, (b) presence of psychiatric disorders, (c) major medical disorders (e.g. diabetes, hypertension, etc.), and (d) any prior diagnosis of infertility or sub-fertility including related conditions such as polycystic ovarian syndrome (PCOS), endometriosis, or pelvic inflammatory disease. Institutional Review Board approval was obtained at each study site and the data coordinating center. All participants provided written informed consent.

The second cohort included a subset of 76 white women from the BioCycle Study, a longitudinal cohort that followed healthy premenopausal women aged 18–44 for up to 2 menstrual cycles (Wactawski-Wende *et al.*, 2009). Participants were recruited from female volunteers in the western New York state region using a variety of methods including advertising in clinical practices and the University at Buffalo student health service, paid advertising in print media, radio and television interviews, notices sent via listserves, and flyers at the university and throughout the region. A total of 969 women inquired about the study, of whom, 449 (46%) expressed interest, 318 (71%) met eligibility, and 276 (87%) were enrolled. Of those who enrolled, 250 women completed two cycles, 9 completed one cycle, and 17 women dropped out prior to completing at least one cycle. The present analysis involves a prospective assessment of 25(OH)D and other markers of vitamin D metabolism and premenstrual symptoms. Women were included if they self-reported cycle lengths between 21 and 35 days for each menstrual cycle during the past 6 months. Exclusion criteria included the following: (a) psychiatric condition requiring medical therapy in the past year (including premenstrual dysphoric disorder), (b) a history or signs of gynecological problems, (c) endometriosis or self-reported diagnosis of PCOS by a physician, (d) use of oral contraceptives during the past 3 months, (e) use of other medications including lipid-lowering drugs, (f) pregnancy in the last 6 months, (g) chronic disease, or (h) a self-reported BMI at screening <18 or $>35\ kg/m^2$. Full details on inclusion and exclusion criteria have been reported elsewhere (Wactawski-Wende *et al.*, 2009). Women in this subsample all had multiple measures of vitamin D metabolism and calcium homeostasis; all were self-reported white race to minimize variability of 25(OH)D by race (Institute of Medicine Committee to Review Dietary Reference Intakes for Vitamin & Calcium, 2011; Harmon *et al.*, 2020). The University at Buffalo Health Sciences Institutional Review Board approved the study and served as the designated Institutional Review Board by the NIH under a reliance agreement. All participants provided written informed consent.

Premenstrual symptomatology assessment

For both cohorts, premenstrual symptoms were assessed via self-report using the same questionnaire (as described below). For EAGeR, the symptoms questionnaire was administered at baseline and queried premenstrual symptoms over the past 12 months, while in BioCycle, the questionnaire was administered prospectively at multiple time points over two menstrual cycles and queried symptoms over the previous week. Specifically, the questionnaire detailed five psychological symptoms (depression or sadness; tension or irritability; anxiety or nervousness; anger, aggression, or short temper; and crying spells) and nine physical symptoms (swelling of hands and/or feet; breast tenderness or fullness; abdominal bloating; lower abdominal cramping; generalized aches and pains; lower backache; headache; fatigue; and insomnia) associated with premenstrual symptoms. Women were asked to rate their individual symptoms during a typical week leading up to menses and a typical week after menses on a scale of: 0 = none, 1 = mild, 2 = moderate, or 3 = severe. A total symptom severity score was calculated by adding the severity ratings for all symptoms (total possible score range 0–42). Scores were calculated for the week prior to menses and the week after menses for comparison. Participants were denoted as either having moderate/severe or mild/none for each individual symptom during the premenstrual week. The total number of moderate/severe symptoms during the premenstrual week was calculated overall and by symptom type.

Biomarker analysis

In both cohorts, serum blood samples were stored at $-80°C$ until analysis. Blood samples for measurement of 25(OH)D were collected at the baseline during menses in EAGeR and during the mid-follicular phase in BioCycle. 25(OH)D concentrations were measured using the 25(OH)D ELISA solid phase sandwich enzyme immunoassay (BioVendor R&D, Ashville, NC, USA) at the University of Minnesota. The interassay laboratory coefficients of variation (CVs) were 15.8% and 13.1% at mean concentrations of 15.5 ng/ml and 41.6 ng/ml, respectively, for lyophilized manufacturer's controls, and 17% for an in-house pooled serum control for EAGeR and 10% for BioCycle. The lower limit of detection was 1.6 ng/ml and all values were above this limit.

In the BioCycle study, additional markers of vitamin D metabolism and calcium homeostasis were measured in blood samples collected during the follicular and ovulatory phases (up to 4 samples per woman). Enzyme immunoassays were used to measure $1,25(OH)_2D$ (EIA Immuno Diagnostics Systems, Fountain Hills, AZ, USA), iPTH (ELISA Abnova, Walnut, CA, USA), and FGF23 (ELISA Kit Kainos Laboratories, Tokyo, Japan, and two-step ELISA using Beckman Coulter Biomek NXp, Beckman Coulter, Inc, Fullerton, CA, USA) at the University of Minnesota. High sensitivity C-Reactive Protein (CRP) was measured using the IMMULITE 2000 chemiluminescent immunoassay at the Kaleida Health Center for Laboratory Medicine, Buffalo, NY, USA. Calcium concentrations were measured using the ion-selective electrode module on the Beckman LX2 automated chemistry analyzer at the Kaleida Health Center for Laboratory Medicine, Buffalo, NY, USA with CVs <5%. Interassay CVs reported by the laboratory for in-house pooled serum controls were 12.2% for $1,25(OH)_2D$, 16.5% for iPTH, and 4.9% for FGF23.

Covariate assessment

At baseline, participants completed questionnaires on demographics (age, race, marital status, education, income), lifestyle factors (smoking, alcohol intake, physical activity), and reproductive health (parity, hormonal contraceptive use, age at menarche, menstrual cycle history, number of prior losses). Height and weight were measured by trained research staff using standardized protocols to calculate BMI. Where possible, standardized questionnaires were utilized. Specifically, physical activity was assessed via the International Physical Activity Questionnaire (IPAQ) (Craig *et al.*, 2003). In BioCycle, depressive

symptoms were assessed using the Center for Epidemiologic Studies Depression Scale (CES-D) (Radloff, 1977) administered at the baseline, and in EAGeR urinary anti-depressant use was measured using the Randox Evidence Investigator 'Drugs of Abuse IV' biochip competitive chemiluminescent immunoassay array (Randox Toxicology, Baltimore, MD, USA) (Sjaarda *et al.*, 2020).

Statistical analysis

Participants were categorized as sufficient (25(OH)D $\geq$ 30.0 ng/ml) or insufficient (25(OH)D $<$ 30.0 ng/ml), based on Endocrine Society guidelines (Holick *et al.*, 2011). Demographic characteristics and reproductive histories were compared by 25(OH)D status and number of moderate/severe symptoms using Student's *t*-tests and Chi-square tests, as appropriate.

Log-binomial regression was used to estimate risk ratios (RR) and 95% CIs for the association between 25(OH)D and risk of individual moderate/severe symptoms during the premenstrual week in each cohort separately. Linear regression was used to estimate β-coefficients and 95% CI for the association between 25(OH)D and the total number of moderate/severe symptoms, and the total number of moderate/severe psychological and physical symptoms in each cohort. All models were adjusted for age, BMI, current smoking status, race, income, physical activity, and season of blood draw. We also compared these results to models that additionally adjust for depression (using the depression scale measured in BioCycle and anti-depressant use in EAGeR), education, alcohol, race (EAGeR only), CRP, and parity. These covariates were chosen a priori based on a review of the prior literature and directed acyclic graph criteria (Greenland *et al.*, 1999; Hernan *et al.*, 2002), as these factors have been associated with both premenstrual symptoms and 25(OH)D insufficiency (Lagunova *et al.*, 2009; Yamamoto *et al.*, 2009; Bertone-Johnson *et al.*, 2010, 2014; Hibler *et al.*, 2016; Rad *et al.*, 2018). Analyses considered both categorical 25(OH)D status (sufficient vs. insufficient) and continuous 25(OH)D concentrations per 10 ng/ml increase. For BioCycle, 25(OH)D serum measurements were assessed in the follicular phase of the first cycle, and premenstrual symptoms were assessed prior to the beginning of the second cycle. This allowed us to assess 25(OH)D status prior to assessment of premenstrual symptoms to prospectively measure their relationship. For EAGeR, 25(OH)D was assessed at the baseline for all study participants and premenstrual symptoms were based on retrospective assessment of the past 12 months. False discovery rate was used to adjust for multiple comparisons.

Associations between premenstrual symptoms and other factors involved in vitamin D metabolism and calcium homeostasis, including 1,25(OH) $_2$D, calcium, FGF23, and iPTH, were evaluated in the BioCycle study. Log-binomial regression was used to estimate RR and 95% CI and models were adjusted for the same factors listed above. The average levels of these factors during the follicular phase were used to evaluate their associations with premenstrual symptoms given that no departures from normality were observed for these factors.

Sensitivity analyses evaluating the potential for associations with the total number of moderate/severe symptoms, and total physical and total psychological symptoms, to be modified by season were also evaluated in the EAGeR cohort, given that insufficiency in the summer months may potentially reflect chronic insufficiency.

Additionally, a sensitivity analysis was conducted to evaluate associations between 25(OH)D and clusters of premenstrual symptoms. Emotional, psychological, physical, and consumption symptom groupings were defined based on patterns identified using factor analysis in prior work by Quintana-Zinn *et al.* (2017) and a score was calculated for each grouping. Scores were based on the sum of symptom severity ratings reported during the premenstrual week (0 = none, 1 = mild, 2 = moderate, 3 = severe). The emotional symptom score included the symptoms: tension or irritability; anger, aggression, short temper; and crying spells. The psychological score included the symptoms: depression or sadness; anxiety or nervousness; and insomnia. The physical score included the symptoms: breast tenderness or fullness; lower abdominal cramping; generalized aches and pains; lower backache; and headache. The consumption score included the symptoms: abdominal bloating and fatigue.

The Hosmer–Lemeshow Goodness of Fit Test was used to assess model fit. SAS version 9.4 (SAS Institute, Cary, NC, USA) was used for all analyses.

Results

25(OH)D concentrations and premenstrual symptoms data were available for 1191 participants in EAGeR and for 76 in BioCycle. Overall, both cohorts were relatively 25(OH)D sufficient with 47% of EAGeR and 46% of BioCycle participants having 25(OH)D levels $\geq$30.0 ng/ml (Table I). Most participants in both the EAGeR and BioCycle studies were white, and the average age in both cohorts was around 28 years. The EAGeR population had a higher mean BMI (26.3 kg/m^2) than the BioCycle population (23.7 kg/m^2). The mean age of menarche and time for cycles to become regular were similar between cohorts. For both cohorts, women with lower BMI and higher income were more likely to be 25(OH)D sufficient. No associations were observed between 25(OH)D status and age, marital status, education, hormonal contraceptive use, age at menarche, menstrual cycle characteristics, and number of previous pregnancy losses. Women in the EAGeR cohort with five or more moderate/severe premenstrual symptoms had higher BMI and CRP levels were more likely to be nulliparous and have selective serotonin reuptake inhibitor exposure at baseline, and less likely to describe their periods as regular during the past 12 months compared to women with fewer symptoms (Supplementary Table SI). Among women in the BioCycle cohort, women with five or more moderate/severe premenstrual symptoms had higher depression scale scores than women with fewer symptoms (Supplementary Table SI).

In both cohorts, abdominal bloating and cramping were the two most common physical symptoms reported (Table II). 25(OH)D insufficiency was associated with increased risk of the total number of moderate/severe physical symptoms and some specific physical symptoms in EAGeR and BioCycle. Specifically, 25(OH)D insufficiency was associated with an increased incidence of breast tenderness or fullness (EAGeR RR 1.27, 95% CI 1.03, 1.55; BioCycle RR 1.37, 95% CI 0.56, 3.32) and generalized aches and pains (EAGeR RR 1.33 95% CI 1.01, 1.78; BioCycle RR 1.36, 95% CI 0.41, 4.45). Point estimates were similar for both cohorts, though confidence intervals were wider in the BioCycle cohort due to the smaller sample size for that cohort. Similar results were observed with the continuous models, as well as after

Table I Baseline characteristics of women in the EAGeR and BioCycle cohorts by serum 25(OH)D concentrations.

	EAGeR				BioCycle				
	Overall	25(OH)D Sufficient (≥ 30 ng/ml)	25(OH)D Insufficient (< 30 ng/ml)	p-value	Overall	Vitamin D Subset	25(OH)D Sufficient (≥ 30 ng/ml)	25(OH)D Insufficient (< 30 ng/ml)	p-value
N	**1191**	555	636		**259**	**76**	35	41	
Age, years	28.7 ± 4.8	28.9 ± 4.8	28.6 ± 4.8	0.36	27.3 ± 8.2	28.4 ± 8.6	28.4 ± 8.6	28.4 ± 8.7	0.99
BMI, kg/m^2	26.3 ± 6.6	24.5 ± 5.1	27.8 ± 7.3	<0.0001	24.1 ± 3.9	23.7 ± 3.5	23.3 ± 3.3	24 ± 3.7	0.36
Race, n (%)				<0.0001					NA
White	1128 (94.7)	542 (97.7)	586 (92.1)		148 (59.2)	76 (100)	35 (100)	41 (100)	
Non-White	63 (5.3)	13 (2.3)	50 (7.9)		102 (40.8)	0 (0)	0 (0)	0 (0)	
Married or living with partner, n (%)	1162 (97.6)	542 (97.7)	620 (97.5)	1	65 (26)	27 (35.5)	15 (42.9)	12 (29.3)	0.24
Education > high school, n (%)	1033 (86.7)	491 (88.5)	542 (85.2)	0.10	226 (87.3)	70 (92.1)	32 (91.4)	38 (92.7)	1
Income, n (%)				0.0004					0.21
≥ $100 000	470 (39.5)	209 (37.7)	261 (41.1)		24 (9.3)	8 (10.5)	3 (8.6)	5 (12.2)	
$75 000–$99 999	147 (12.4)	84 (15.1)	63 (9.9)		45 (17.5)	13 (17.1)	9 (25.7)	4 (9.8)	
$40 000–$74 999	174 (14.6)	99 (17.8)	75 (11.8)		72 (28)	29 (38.2)	15 (42.9)	14 (34.1)	
$20 000–$39 999	307 (25.8)	128 (23.1)	179 (28.2)		61 (23.7)	15 (19.7)	5 (14.3)	10 (24.4)	
≤ $19 999	92 (7.7)	35 (6.3)	57 (9.0)		55 (21.4)	11 (14.5)	3 (8.6)	8 (19.5)	
Current smokers, n (%)	148 (12.5)	69 (12.5)	79 (12.5)	1		5 (6.6)	3 (8.6)	2 (4.9)	0.66
Parity, n (%)				0.09					0.21
Nulliparous	550 (46.2)	271 (48.8)	279 (43.9)		187 (73.9)	53 (70.7)	22 (62.9)	31 (77.5)	
Parous (1 or 2 prior live births)	641 (53.8)	284 (51.2)	357 (56.1)		66 (26.1)	22 (29.3)	13 (37.1)	9 (22.5)	
Ever used hormonal contraception/meds, n (%)	907 (80.1)	435 (81.6)	472 (78.8)	0.26	140 (54.9)	45 (60.0)	22 (62.9)	23 (57.5)	0.81
Age at menarche, years	12.7 ± 1.5	12.8 ± 1.5	12.7 ± 1.5	0.21	12.5 ± 1.2	12.5 ± 1.1	12.7 ± 1.1	12.4 ± 1.0	0.21
Age for periods to become regular, year	14.3 ± 4.5	14.4 ± 4.5	14.3 ± 4.5	0.85	14.8 ± 2.9	15 ± 3.0	14.9 ± 2.6	15.1 ± 3.4	0.76
Usual menstrual bleeding, days	5.1 ± 1.8	5.0 ± 1.3	5.1 ± 2.1	0.52	5.1 ± 1.1	5.3 ± 0.9	5.3 ± 1.0	5.4 ± 0.9	0.58
Usual cycle length over last 12 months, days	29.2 ± 4	29.2 ± 4.1	29.3 ± 3.9	0.84	28.4 ± 2.2	28.3 ± 2.4	28.2 ± 2.6	28.3 ± 2.2	0.85
Shortest cycle length in last 12 months, days	26.2 ± 4.6	26 ± 4.8	26.3 ± 4.3	0.34	25.3 ± 3.7	25.3 ± 3.8	24.8 ± 4	25.6 ± 3.7	0.37
Longest cycle in last 12 months, days	33.9 ± 9.8	33.4 ± 8.5	34.3 ± 11	0.17	31.3 ± 6.2	31.6 ± 5.2	30.9 ± 2.8	32.2 ± 6.6	0.30
Describe regularity of period in the last 12 months, n (%)				0.05					1
Yes	939 (82.9)	439 (81.6)	500 (84)		249 (96.1)	70 (92.1)	33 (94.3)	37 (90.2)	
No	138 (12.2)	77 (14.3)	61 (10.3)		6 (2.3)	5 (6.6)	2 (5.7)	3 (7.3)	
Don't Know	56 (4.9)	22 (4.1)	34 (5.7)		4 (1.5)	1 (1.3)	0 (0)	1 (2.4)	
Number of previous pregnancy losses, n (%)				0.85					0.53
0					43 (55.8)	15 (62.5)	9 (69.2)	6 (54.5)	
1	799 (67.1)	425 (66.8)	374 (67.4)		22 (28.6)	8 (33.3)	4 (30.8)	4 (36.4)	

(continued)

Table I Continued

	EAGeR				BioCycle			
	Overall	25(OH)D Sufficient (≥ 30 ng/ml)	25(OH)D Insufficient (< 30 ng/ml)	p-value	Overall	Vitamin D Subset	25(OH)D Sufficient (≥ 30 ng/ml)	25(OH)D Insufficient (< 30 ng/ml)	p-value
2	392 (32.9)	211 (33.2)	181 (32.6)		10 (13)	1 (4.2)	0 (0)	1 (9.1)	
3					2 (2.6)	0 (0)	0 (0)	0 (0)	
SSRI exposure at baseline, n (%)									
Yes	168 (14.2)	89 (14.1)	79 (14.3)	0.93					
No	1015 (85.8)	543 (85.9)	472 (85.7)						
Depression scale					4.7 ± 1.4	4.7 ± 1.4	4.5 ± 1.4	4.9 ± 1.4	0.21
Alcohol intake, n(%)				0.002					0.05
Often	26 (2.2)	14 (2.2)	12 (2.2)		19 (7.3)	7 (9.2)	4 (11.4)	3 (7.3)	
Sometimes	368 (31.3)	168 (26.8)	200 (36.4)		153 (59.1)	56 (73.7)	29 (82.9)	27 (65.9)	
Never	782 (66.5)	445 (71)	337 (61.4)		87 (33.6)	13 (17.1)	2 (5.7)	11 (26.8)	
CRP, mg/L	1.14 ± 2.91	1.32 ± 2.93	0.97 ± 2.82	<0.001	0.63 ± 2.65	0.69 ± 2.30	0.67 ± 2.33	0.71 ± 2.30	0.75

25(OH)D, 25-hydroxyvitamin D; CRP, high sensitivity c-reactive protein; EAGeR, Effects of Aspirin in Gestation and Reproduction; SSRI, selective serotonin reuptake inhibitors.

additional adjustment for depression, education, alcohol, race (EAGeR only), CRP, and parity. Associations with the total number of moderate/severe physical symptoms remained after adjustment for the false discovery rate, though for individual symptoms the associations were no longer statistically significant after adjustment.

In both cohorts, the most common psychological symptom was tension or irritability, followed by anger, aggression, short temper, and anxiety or nervousness (Table III). 25(OH)D insufficiency was not associated with an increase in the number of moderate/severe psychological symptoms or individual psychological premenstrual symptoms in either cohort. Similar results were observed with both adjusted models.

Other markers related to vitamin D metabolism and calcium homeostasis in the BioCycle study, including iPTH, calcium, FGF23, and 1,25(OH) $_2$D, were not associated with physical or psychological premenstrual symptoms (Tables IV and V). Results of the Hosmer–Lemeshow Goodness of Fit test did not provide evidence of poor model fit.

In results stratified by season, we observed that 25(OH)D insufficiency in the summer was associated with increased risk of total number of moderate/severe premenstrual symptoms, as well as total physical symptoms in the EAGeR cohort (Supplementary Table SII). A suggestion of an increased risk of total psychological symptoms was observed in the summer as well. Associations were not observed in the other seasons.

25(OH)D insufficiency was associated with higher physical symptom cluster scores, though results were attenuated after adjustment for confounders (Supplementary Table SIII). No associations were observed between 25(OH)D insufficiency or continuous 25(OH)D concentrations and emotional, psychological, or consumption symptom cluster scores (Supplementary Table SIII).

Discussion

25(OH)D insufficiency was associated with increased risk of specific physical premenstrual symptoms, namely breast fullness/tenderness and generalized aches and pain, though not with psychological symptoms. Currently, treatments for premenstrual symptoms such as hormonal contraceptives may result in side effects and few non-hormonal treatments are available for symptom relief (Direkvand-Moghadam *et al.*, 2014). These findings highlight a potentially modifiable factor for improving specific premenstrual symptoms that affect approximately 50% of the female population.

Our findings are consistent with other studies of the relationship between 25(OH)D and premenstrual symptoms. Though studies utilizing dietary intake have suggested associations between vitamin D and incident PMS in both college-age women (Bertone-Johnson *et al.*, 2010) and women older than 30 in the Nurses' Health Study II (Bertone-Johnson *et al.*, 2005), measured 25(OH)D concentrations were not associated with PMS in either cohort women (Bertone-Johnson *et al.*, 2014). However, in the NHS2 cohort, plasma 25(OH)D was associated with specific physical symptoms, including breast tenderness, fatigue, diarrhea/constipation, and depression (Bertone-Johnson *et al.*, 2014). These previous studies support our findings of an association between serum 25(OH)D and certain premenstrual symptoms, though not the overall syndrome itself. Our findings, particularly among

Table II Association between serum 25(OH)D concentrations and moderate/severe physical premenstrual symptoms during the premenstrual week in two cohorts of reproductive age women with regular cycles: EAGeR and BioCycle.

	EAGeR			BioCycle		
	25(OH)D sufficient (≥ 30 ng/ml)	25(OH)D insufficient (< 30 ng/ml)	25(OH)D (per 10 ng/ml)	25(OH)D sufficient (≥ 30 ng/ml)	25(OH)D insufficient (< 30 ng/ml)	25(OH)D (per 10 ng/ml)
Total number of moderate/severe physical symptoms						
Mean ± SD	1.9 ± 2.0	2.3 ± 2.3		2.0 ± 1.8	2.0 ± 1.8	
Unadjusted β (95% CI)	Ref	**0.42 (0.17, 0.67)**	**−0.15 (−0.25, −0.05)**	Ref	0.03 (−0.84, 0.89)	−0.10 (−0.59, 0.39)
Adjusted 1: β (95% CI)	Ref	**0.31 (0.06, 0.51)**	−0.10 (−0.20, 0.01)	Ref	−0.06 (−1.01, 0.89)	−0.13 (−0.69, 0.43)
Adjusted 2: β (95% CI)	Ref	0.25 (0.00, 0.51)	**−0.15 (−0.25, −0.05)***	Ref	−0.12 (−1.14, 0.90)	−0.03 (−0.63, 0.56)
Swelling of hands or feet						
Prevalence	17 (3.1)	32 (5.4)		1 (2.9)	4 (10.3)	
Unadjusted RR (95% CI)	Ref	1.75 (0.96, 3.18)	0.76 (0.56, 1.02)	Ref	3.49 (0.39, 30.84)	0.67 (0.23, 1.96)
Adjusted 1: RR (95% CI)	Ref	1.67 (0.87, 3.23)	0.78 (0.56, 1.08)	Ref	2.96 (0.34, 26.04)	0.31 (0.05, 1.97)
Adjusted 2: RR (95% CI)	Ref	1.6 (0.84, 3.07)	0.81 (0.58, 1.12)	Ref	1.89 (0.15, 23.91)	0.56 (0.12, 2.7)
Breast tenderness or fullness						
Prevalence	127 (23.5)	186 (31.2)		7 (21.2)	11 (28.2)	
Unadjusted RR (95% CI)	Ref	**1.33 (1.10, 1.61)***	**0.87 (0.79, 0.95)***	Ref	1.33 (0.57, 3.08)	0.93 (0.60, 1.46)
Adjusted 1: RR (95% CI)	Ref	**1.27 (1.03, 1.55)**	**0.89 (0.81, 0.98)**	Ref	1.37 (0.56, 3.32)	0.88 (0.54, 1.45)
Adjusted 2: RR (95% CI)	Ref	**1.30 (1.05, 1.61)**	**0.89 (0.80, 0.99)**	Ref	2.02 (0.49, 8.33)	0.76 (0.38, 1.48)
Abdominal bloating						
Prevalence	169 (31.2)	219 (36.6)		11 (32.4)	18 (46.2)	
Unadjusted RR (95% CI)	Ref	1.17 (0.995, 1.38)	0.94 (0.88, 1.01)	Ref	1.43 (0.78, 2.61)	0.93 (0.66, 1.30)
Adjusted 1: RR (95% CI)	Ref	1.17 (0.98, 1.39)	0.95 (0.88, 1.03)	Ref	1.31 (0.63, 2.74)	0.95 (0.64, 1.40)
Adjusted 2: RR (95% CI)	Ref	1.14 (0.96, 1.37)	0.95 (0.87, 1.03)	Ref	1.61 (0.44, 5.85)	0.84 (0.46, 1.51)
Lower abdominal cramping						
Prevalence	178 (32.9)	241 (40.3)		16 (47.1)	16 (41)	
Unadjusted RR (95% CI)	Ref	**1.22 (1.05, 1.43)***	0.93 (0.86, 1.00)	Ref	0.87 (0.51, 1.48)	1.11 (0.85, 1.45)
Adjusted 1: RR (95% CI)	Ref	1.13 (0.96, 1.33)	0.97 (0.90, 1.04)	Ref	0.89 (0.48, 1.64)	1.05 (0.75, 1.45)
Adjusted 2: RR (95% CI)	Ref	1.13 (0.95, 1.33)	0.96 (0.89, 1.04)	Ref	0.96 (0.40, 2.29)	1.10 (0.64, 1.88)
Generalized aches and pains						
Prevalence	73 (13.5)	122 (20.4)		4 (11.8)	6 (15.8)	
Unadjusted RR (95% CI)	Ref	**1.52 (1.16, 1.98)***	**0.81 (0.71, 0.93)***	Ref	1.34 (0.40, 4.45)	0.65 (0.32, 1.32)
Adjusted 1: RR (95% CI)	Ref	**1.33 (1.01, 1.78)**	**0.87 (0.76, 0.99)**	Ref	1.36 (0.41, 4.45)	0.60 (0.27, 1.36)
Adjusted 2: RR (95% CI)	Ref	1.25 (0.94, 1.67)	**0.87 (0.75, 1.00)**	Ref	1.27 (0.14, 11.41)	0.65 (0.16, 2.68)
Lower backache						
Prevalence	112 (20.7)	157 (26.3)		10 (29.4)	7 (17.9)	
Unadjusted RR (95% CI)	Ref	**1.27 (1.03, 1.57)**	0.95 (0.87, 1.05)	Ref	0.61 (0.26, 1.45)	0.97 (0.61, 1.55)
Adjusted 1: RR (95% CI)	Ref	1.19 (0.95, 1.48)	0.99 (0.90, 1.09)	Ref	0.42 (0.15, 1.12)	1.14 (0.67, 1.94)
Adjusted 2: RR (95% CI)	Ref	1.12 (0.89, 1.41)	1.02 (0.92, 1.12)	Ref	0.48 (0.13, 1.72)	1.06 (0.57, 1.97)
Headache						
Prevalence	141 (26.0)	153 (25.8)		7 (20.6)	7 (17.9)	
Unadjusted RR (95% CI)	Ref	0.99 (0.81, 1.21)	0.97 (0.89, 1.06)	Ref	0.87 (0.33, 2.27)	0.99 (0.58, 1.67)
Adjusted 1: RR (95% CI)	Ref	0.94 (0.76, 1.16)	1.00 (0.92, 1.09)	Ref	0.96 (0.39, 2.32)	1.10 (0.58, 2.09)
Adjusted 2: RR (95% CI)	Ref	0.91 (0.73, 1.13)	1.01 (0.93, 1.11)	Ref	0.73 (0.20, 2.68)	1.13 (0.55, 2.34)
Fatigue						
Prevalence	152 (28.1)	198 (33.2)		10 (29.4)	8 (20.5)	
Unadjusted RR (95% CI)	Ref	1.18 (0.99, 1.41)	0.93 (0.85, 1.01)	Ref	0.70 (0.31, 1.59)	1.05 (0.67, 1.66)
Adjusted 1: RR (95% CI)	Ref	1.09 (0.90, 1.31)	0.97 (0.89, 1.05)	Ref	0.82 (0.32, 2.10)	0.88 (0.49, 1.60)
Adjusted 2: RR (95% CI)	Ref	1.07 (0.89, 1.3)	0.97 (0.89, 1.06)	Ref	0.78 (0.26, 2.36)	0.98 (0.54, 1.79)

(continued)

Table II Continued

	EAGeR			BioCycle		
	25(OH)D sufficient (≥ 30 ng/ml)	25(OH)D insufficient (< 30 ng/ml)	25(OH)D (per 10 ng/ml)	25(OH)D sufficient (≥ 30 ng/ml)	25(OH)D insufficient (< 30 ng/ml)	25(OH)D (per 10 ng/ml)
Insomnia						
Prevalence	46 (8.5)	63 (10.5)		1 (3)	1 (2.6)	
Unadjusted RR (95% CI)	Ref	1.24 (0.86, 1.78)	0.93 (0.79, 1.10)	Ref	0.85 (0.05, 13.65)	0.36 (0.05, 2.75)
Adjusted 1: RR (95% CI)	Ref	1.14 (0.78, 1.67)	0.97 (0.83, 1.15)	Ref	0.57 (0.03, 10.41)	0.18 (0.00, 20.95)
Adjusted 2: RR (95% CI)	Ref	1.17 (0.79, 1.73)	0.99 (0.84, 1.18)	Ref	0.69 (0, 114.24)	0.98 (0.05, 18.97)

25(OH)D, 25-hydroxyvitamin D; EAGeR, Effects of Aspirin in Gestation and Reproduction; Ref, Reference; RR, risk ratio.
Bold results are significant at $\alpha = 0.05$.
*Indicates comparisons that remained after adjustment for false discovery rate.
Adjusted 1 model controls for age, BMI, smoking status, race, income, exercise, season of blood draw.
Adjusted 2 model controls for the variables in Adjusted 1 as well as depression (BioCycle) or antidepressant use (EAGeR), education, alcohol, race (EAGeR only), CRP (high sensitivity c-reactive protein), and parity.

Biocycle participants, extend this prior research, suggesting that 25(OH)D may be associated with cycle-specific incidence of premenstrual symptoms, though we had limited power to adequately assess this relationship.

Additionally, other trials have also shown that 25(OH)D supplementation may be associated with improved dysmenorrhea (Lasco *et al.*, 2012). The role of 25(OH)D on specific physical symptoms in NHS2 (Bertone-Johnson *et al.*, 2014) is consistent with our findings for 25(OH)D insufficiency and increased physical symptoms in EAGeR, and of a similar magnitude and direction for BioCycle though not statistically significant. Our work extends prior work by utilizing two different cohorts, one that is able to evaluate short term associations between serum 25(OH)D and premenstrual symptoms in the subsequent cycle; and a second larger cohort assesses associations between serum 25(OH)D and a pattern of premenstrual symptoms over the last twelve months. Inclusion of both cohorts allows for a more comprehensive approach to assess the relationship between 25(OH)D insufficiency and individual premenstrual symptoms among women with self-reported regular cycles, although it is important to note that both cohorts are primarily of self-reported white race and more diverse cohorts are needed to improve generalizability. Further, results should be interpreted cautiously given that associations with individual symptoms were no longer statistically significant after adjustment for false discovery rate.

The mechanisms underlying the observed associations between 25(OH)D and physical premenstrual symptoms are likely multifactorial. We found that 25(OH)D was associated with breast tenderness and general aches and pains, which may be related to inflammatory pathways that increase pain signaling (Tague and Smith, 2011) though specific mechanisms are not well understood and associations with abdominal cramping were imprecise. In regard to other physical symptoms, 25(OH)D has also been hypothesized to improve dysmenorrhea via a reduction in proinflammatory cytokines (Lasco *et al.*, 2012; Thys-Jacobs, 2000). Moreover, vitamin D has been used to treat aromatase inhibitor musculoskeletal symptoms, which are a cluster of musculoskeletal symptoms caused by estrogen dysregulation, with symptomatology similar to PMS, suggesting similar pathophysiologic mechanisms (Waltman *et al.*, 2009; Khan *et al.*, 2010; Prieto-Alhambra *et al.*, 2011; Rastelli *et al.*, 2011; Khan *et al.*, 2012; Singer *et al.*, 2014; Shapiro *et al.*, 2016). Results also suggested that 25(OH)D insufficiency in the summer may be particularly relevant for premenstrual symptoms, which may potentially be a result of insufficiency in the summer being more likely to reflect chronic insufficiency.

Other markers of vitamin D metabolism and calcium homeostasis have also been linked to PMS in other studies (Thys-Jacobs *et al.*, 2000). Calciotropic hormones including iPTH, calcium, FGF23, and 1,25(OH)$_2$D have been suggested to play a role as hypocalcemia is known to cause symptoms that are similar to those that occur in PMS, such as muscle cramps, irritability, anxiety, and fatigue (Schafer, 2016). Further, calcium has known physiological effects on muscle relaxation and contractility (Noble *et al.*, 2009). While some evidence suggests that women with luteal phase symptomatology have an underlying calcium dysregulation with a secondary hyperparathyroidism and 25(OH)D deficiency (Morales *et al.*, 2008), our findings do not support an association of iPTH, calcium, FGF23, and 1,25(OH)$_2$D concentrations with premenstrual symptoms; however, our sample size was limited.

This study has several important strengths and limitations. Our analysis included two cohorts of women with regular cycles, one which was a cross-sectional assessment (EAGeR), and one which was prospective (BioCycle) which enabled us to assess the consistency of associations between 25(OH)D and specific premenstrual symptoms. Of note, reverse causality is a potential concern for results from the EAGeR study as both 25(OH)D and individual premenstrual symptoms over the past year were assessed at baseline. Our assessment of serum 25(OH)D concentration is a strength as dietary assessments do not capture vitamin D from all sources and serum concentrations are an accurate biological reflection of 25(OH)D status regardless of source (Takeuchi *et al.*, 1995; Thomas *et al.*, 1998). BioCycle also allowed us to evaluate 1,25(OH)$_2$D, iPTH, calcium, and FGF23 which let us further investigate associations with multiple markers involved in vitamin D metabolism and calcium homeostasis on premenstrual symptom outcomes. Another strength of our BioCycle assessment was a short duration between the serum 25(OH)D and premenstrual symptom score assessment (approximately 2 weeks), which better

Table III Serum 25(OH)D concentrations and risk of moderate/severe psychological premenstrual symptoms during the premenstrual week in two cohorts of reproductive age women with regular cycles: EAGeR and BioCycle.

	EAGeR			BioCycle		
	25(OH)D sufficient (≥ 30 ng/ml)	25(OH)D insufficient (< 30 ng/ml)	25(OH)D (per 10 ng/ml)	25(OH)D sufficient (≥ 30 ng/ml)	25(OH)D insufficient (< 30 ng/ml)	25(OH)D (per 10 ng/ml)
Total number of moderate/severe psychological symptoms						
Mean ± SD	1.2 ± 1.5	1.2 ± 1.5		0.7 ± 1.1	1.2 ± 1.3	
Unadjusted β (95% CI)	Ref	0.07 (−0.11, 0.25)	−0.01 (−0.08, 0.06)	Ref	0.40 (−0.19, 0.98)	−0.23 (−0.57, 0.10)
Adjusted 1: β (95% CI)	Ref	0.02 (−0.17, 0.21)	0.01 (−0.07, 0.09)	Ref	0.34 (−0.25, 0.93)	−0.24 (−0.59, 0.11)
Adjusted 2: β (95% CI)	Ref	0.03 (−0.16, 0.22)	0.02 (−0.06, 0.10)	Ref	0.26 (−0.27, 0.80)	−0.22 (−0.53, 0.09)
Depression or sadness						
Prevalence	91 (16.8)	107 (17.9)		3 (8.8)	5 (12.8)	
Unadjusted RR (95% CI)	Ref	1.06 (0.83, 1.37)	1.01 (0.92, 1.12)	Ref	1.45 (0.37, 5.77)	0.58 (0.25, 1.35)
Adjusted 1: RR (95% CI)	Ref	1.07 (0.82, 1.40)	0.99 (0.90, 1.12)	Ref	1.34 (0.29, 6.16)	0.45 (0.12, 1.63)
Adjusted 2: RR (95% CI)	Ref	1.10 (0.84, 1.44)	1.00 (0.89, 1.12)	Ref	1.42 (0.10, 19.7)	0.63 (0.11, 3.67)
Tension or irritability						
Prevalence	215 (39.8)	246 (41.3)		8 (23.5)	13 (33.3)	
Unadjusted RR (95% CI)	Ref	1.04 (0.90, 1.20)	0.99 (0.93, 1.05)	Ref	1.42 (0.66, 3.04)	0.81 (0.53, 1.24)
Adjusted 1: RR (95% CI)	Ref	1.00 (0.86, 1.16)	1.00 (0.94, 1.07)	Ref	1.41 (0.61, 3.29)	0.68 (0.39, 1.20)
Adjusted 2: RR (95% CI)	Ref	0.98 (0.84, 1.14)	1.02 (0.95, 1.09)	Ref	1.64 (0.58, 4.66)	0.59 (0.28, 1.23)
Anxiety or nervousness						
Prevalence	97 (17.9)	116 (19.4)		5 (14.7)	10 (25.6)	
Unadjusted RR (95% CI)	Ref	1.09 (0.85, 1.38)	1.03 (0.93, 1.14)	Ref	1.74 (0.65, 4.68)	0.73 (0.42, 1.28)
Adjusted 1: RR (95% CI)	Ref	1.06 (0.82, 1.36)	1.04 (0.94, 1.16)	Ref	2.93 (0.79, 10.79)	0.50 (0.19, 1.30)
Adjusted 2: RR (95% CI)	Ref	1.04 (0.80, 1.34)	1.06 (0.95, 1.19)	Ref	2.14 (0.22, 20.85)	0.56 (0.14, 2.26)
Anger, aggression, short temper						
Prevalence	154 (28.5)	171 (28.6)		6 (18.2)	12 (30.8)	
Unadjusted RR (95% CI)	Ref	1.00 (0.84, 1.21)	0.96 (0.89, 1.04)	Ref	1.69 (0.70, 4.07)	0.70 (0.43, 1.16)
Adjusted RR (95% CI)	Ref	0.93 (0.77, 1.13)	1.00 (0.92, 1.08)	Ref	2.19 (0.78, 6.13)	0.64 (0.33, 1.24)
Adjusted 2: RR (95% CI)	Ref	0.92 (0.76, 1.13)	1.01 (0.92, 1.10)	Ref	1.18 (0.22, 6.19)	0.84 (0.30, 2.39)
Crying spells						
Prevalence	75 (13.9)	100 (16.8)		2 (5.9)	3 (7.7)	
Unadjusted RR (95% CI)	Ref	1.21 (0.92, 1.60)	0.96 (0.85, 1.09)	Ref	1.31 (0.23, 7.59)	1.43 (0.58, 3.53)
Adjusted 1: RR (95% CI)	Ref	1.11 (0.83, 1.48)	1.00 (0.88, 1.13)	Ref	2.01 (0.18, 22.43)	1.18 (0.25, 5.50)
Adjusted 2: RR (95% CI)	Ref	1.24 (0.92, 1.68)	0.98 (0.86, 1.12)	Ref	0.94 (0.08, 11.57)	1.31 (0.24, 7.08)

25(OH)D, 25-hydroxyvitamin D; EAGeR, Effects of Aspirin in Gestation and Reproduction; Ref, Reference; RR, risk ratio.
Bold results are significant at $\alpha = 0.05$.
*Indicates comparisons that remained after adjustment for false discovery rate.
Adjusted 1 model controls for age, BMI, smoking status, race, income, exercise, and season of blood draw.
Adjusted 2 model controls for the variables in Adjusted 1 as well as depression (BioCycle) or antidepressant use (EAGeR), education, alcohol, race (EAGeR only), CRP (high sensitivity c-reactive protein), and parity.

captures exposure assessment in a time period proximal to individual premenstrual symptoms compared with other studies that had longer periods of time between exposure and outcome assessment. This is particularly important due to seasonal changes in 25(OH)D and hormones which may impact premenstrual symptom outcomes. Premenstrual symptom score assessment for both cohorts was done by self-report, though it is important to note that this is a widely used and accepted form of assessment of milder symptoms of individual premenstrual symptoms as PMS is rarely formally diagnosed by a physician. Further, though we were able to account for many potential confounding factors, information on social support and symptom sensitivity was not available so that the results have the potential for residual confounding by these and other factors, and covariate information on demographics and specific lifestyle and reproductive health covariates were not assessed using standardized instruments.

A limitation is that participants in both studies were primarily of white race, and future studies in more diverse populations are needed. Moreover, this study was limited to women with self-reported regular cycles, though women who were and were not attempting to conceive

Table IV Association between markers of vitamin D metabolism and moderate/severe physical premenstrual symptoms during the premenstrual week in the BioCycle study.

BioCycle study	iPTH (continuous) per 5 pg/ml	1,25-dihydroxyvitamin D (continuous) per 10 pmol/l	FGF23 (continuous) per 5 pg/ml	Calcium (continuous) per 0.1 mg/dl
Total number of moderate/severe physical symptoms				
Unadjusted β (95% CI)	0.06 (−0.10, 0.22)	0.02 (−0.15, 0.19)	0.02 (−0.13, 0.17)	0.02 (−0.10, 0.13)
Adjusted 1: β (95% CI)	−0.01 (−0.18, 0.16)	−0.06 (−0.25, 0.13)	0.03 (−0.14, 0.20)	0.08 (−0.05, 0.20)
Adjusted 2: β (95% CI)	−0.06 (−0.23, 0.12)	−0.08 (−0.27, 0.12)	0.00 (−0.18, 0.19)	0.09 (−0.03, 0.21)
Swelling of hands or feet				
Unadjusted RR (95% CI)	1.23 (0.96, 1.59)	0.89 (0.62, 1.26)	0.97 (0.70, 1.35)	0.89 (0.72, 1.10)
Adjusted 1: RR (95% CI)	1.26 (0.72, 2.21)	0.86 (0.50, 1.49)	1.32 (0.63, 2.76)	1.08 (0.88, 1.33)
Adjusted 2: RR (95% CI)	1.04 (0.58, 1.88)	0.86 (0.50, 1.49)	1.00 (0.52, 1.91)	1.13 (0.87, 1.47)
Breast tenderness or fullness				
Unadjusted RR (95% CI)	1.09 (0.96, 1.24)	1.00 (0.86, 1.17)	1.11 (0.96, 1.30)	0.96 (0.87, 1.06)
Adjusted 1: RR (95% CI)	1.07 (0.92, 1.25)	0.92 (0.73, 1.16)	1.01 (0.80, 1.28)	0.96 (0.85, 1.07)
Adjusted 2: RR (95% CI)	1.08 (0.88, 1.33)	0.90 (0.69, 1.18)	1.02 (0.73, 1.43)	0.99 (0.88, 1.10)
Abdominal bloating				
Unadjusted RR (95% CI)	0.94 (0.83, 1.06)	0.98 (0.88, 1.09)	0.96 (0.85, 1.10)	1.03 (0.96, 1.11)
Adjusted 1: RR (95% CI)	0.90 (0.78, 1.04)	0.98 (0.84, 1.14)	0.94 (0.80, 1.10)	1.05 (0.97, 1.14)
Adjusted 2: RR (95% CI)	0.90 (0.76, 1.07)	0.90 (0.71, 1.12)	0.89 (0.71, 1.11)	1.04 (0.96, 1.14)
Lower abdominal cramping				
Unadjusted RR (95% CI)	1.03 (0.95, 1.12)	1.02 (0.93, 1.11)	1.08 (0.98, 1.18)	1.07 (1.00, 1.14)
Adjusted 1: RR (95% CI)	1.01 (0.90, 1.15)	1.00 (0.87, 1.15)	1.08 (0.87, 1.35)	**1.09 (1.00, 1.18)***
Adjusted 2: RR (95% CI)	1.01 (0.85, 1.21)	0.97 (0.79, 1.19)	1.09 (0.78, 1.52)	1.09 (1.00, 1.19)
Generalized aches and pains				
Unadjusted RR (95% CI)	0.9 (0.70, 1.15)	1.08 (0.88, 1.33)	0.99 (0.81, 1.21)	1.04 (0.92, 1.17)
Adjusted 1: RR (95% CI)	0.82 (0.63, 1.07)	1.07 (0.82, 1.41)	0.99 (0.78, 1.26)	1.07 (0.94, 1.21)
Adjusted 2: RR (95% CI)	0.82 (0.63, 1.07)	0.99 (0.62, 1.58)	1.03 (0.69, 1.54)	1.08 (0.94, 1.23)
Lower backache				
Unadjusted RR (95% CI)	1.05 (0.91, 1.20)	1.06 (0.09, 1.25)	1.03 (0.90, 1.18)	1.04 (0.94, 1.15)
Adjusted 1: RR (95% CI)	1.03 (0.88, 1.22)	1.07 (0.88, 1.31)	1.09 (0.89, 1.33)	1.07 (0.96, 1.19)
Adjusted 2: RR (95% CI)	0.99 (0.81, 1.21)	1.04 (0.82, 1.30)	1.00 (0.77, 1.29)	1.06 (0.95, 1.18)
Headache				
Unadjusted RR (95% CI)	1.07 (0.93, 1.22)	0.95 (0.79, 1.14)	0.98 (0.84, 1.15)	0.94 (0.84, 1.05)
Adjusted 1: RR (95% CI)	1.05 (0.89, 1.23)	0.92 (0.74, 1.15)	1.10 (0.93, 1.31)	0.98 (0.87, 1.11)
Adjusted 2: RR (95% CI)	0.97 (0.79, 1.20)	0.96 (0.71, 1.29)	1.05 (0.81, 1.36)	1.03 (0.90, 1.17)
Fatigue				
Unadjusted RR (95% CI)	1.04 (0.91, 1.19)	1.01 (0.87, 1.18)	0.93 (0.79, 1.10)	0.98 (0.89, 1.07)
Adjusted 1: RR (95% CI)	1.01 (0.84, 1.21)	0.98 (0.79, 1.20)	0.88 (0.71, 1.08)	1.03 (0.93, 1.14)
Adjusted 2: RR (95% CI)	0.94 (0.72, 1.22)	0.92 (0.67, 1.26)	0.77 (0.57, 1.04)	1.03 (0.93, 1.15)
Insomnia				
Unadjusted RR (95% CI)	0.95 (0.61, 1.49)	1.2 (0.77, 1.87)	0.82 (0.48, 1.41)	1.02 (0.83, 1.26)
Adjusted 1: RR (95% CI)	0.92 (0.48, 1.75)	1.00 (0.64, 1.54)	0.93 (0.49, 1.77)	1.14 (0.90, 1.45)
Adjusted 2: RR (95% CI)	1.07 (0.44, 2.63)	0.94 (0.49, 1.83)	0.97 (0.38, 2.53)	1.06 (0.82, 1.36)

FGF23, fibroblast growth factor 23; iPTH, intact parathyroid hormone; RR, risk ratio.
Bold results are significant at $\alpha = 0.05$.
*Indicates comparisons that survived adjustment for false discovery rate.
Adjusted 1 model controls for age, BMI, smoking status, race, income, exercise, season of blood draw.
Adjusted 2 model controls for the variables in Adjusted 1 as well as depression, education, alcohol, CRP (high sensitivity c-reactive protein), and parity.

Table V Markers of vitamin D metabolism and risk of moderate/severe psychological premenstrual symptoms during the premenstrual week in the BioCycle study.

	iPTH (continuous) per 5 pg/ml	1,25-Dihydroxyvitamin D (continuous) Per 10 pmol/l	FGF23 (continuous) per 5 pg/ml	Calcium (continuous) per 0.1 mg/dl
Total number of moderate/severe psychological symptoms				
Unadjusted β (95% CI)	0.07 (−0.03, 0.18)	−0.07 (−0.18, 0.04)	−0.04 (−0.14, 0.06)	−0.01 (−0.09, 0.06)
Adjusted 1: β (95% CI)	0.08 (−0.03, 0.19)	−0.11 (−0.23, 0.01)	−0.01 (−0.12, 0.10)	0.01 (−0.07, 0.09)
Adjusted 2: β (95% CI)	0.06 (−0.03, 0.15)	−0.08 (−0.18, 0.01)	0.00 (−0.09, 0.09)	0.03 (−0.02, 0.08)
Depression or sadness				
Unadjusted RR (95% CI)	1.06 (0.85, 1.34)	1.02 (0.79, 1.33)	0.92 (0.70, 1.22)	0.97 (0.84, 1.11)
Adjusted 1: RR (95% CI)	1.11 (0.82, 1.48)	1.00 (0.76, 1.33)	0.99 (0.68, 1.43)	0.98 (0.85, 1.14)
Adjusted 2: RR (95% CI)	0.89 (0.48, 1.67)	1.00 (0.76, 1.33)	0.93 (0.46, 1.88)	0.96 (0.75, 1.22)
Tension or irritability				
Unadjusted RR (95% CI)	1.06 (0.95, 1.18)	0.89 (0.79, 1.01)	0.94 (0.82, 1.08)	0.99 (0.91, 1.08)
Adjusted 1: RR (95% CI)	1.10 (0.90, 1.35)	0.85 (0.70, 1.03)	0.97 (0.80, 1.16)	1.04 (0.95, 1.15)
Adjusted 2: RR (95% CI)	1.12 (0.88, 1.43)	0.81 (0.59, 1.10)	0.92 (0.73, 1.17)	1.09 (0.98, 1.23)
Anxiety or nervousness				
Unadjusted RR (95% CI)	1.06 (0.91, 1.22)	0.93 (0.77, 1.12)	1.03 (0.89, 1.18)	1.02 (0.91, 1.13)
Adjusted 1: RR (95% CI)	1.23 (0.93, 1.63)	0.86 (0.70, 1.06)	0.92 (0.74, 1.14)	0.98 (0.87, 1.12)
Adjusted 2: RR (95% CI)	1.20 (0.72, 2.00)	0.79 (0.51, 1.23)	1.09 (0.83, 1.44)	1.00 (0.85, 1.17)
Anger, aggression, short temper				
Unadjusted RR (95% CI)	1.08 (0.96, 1.22)	0.91 (0.79, 1.06)	0.90 (0.77, 1.05)	0.97 (0.88, 1.07)
Adjusted RR (95% CI)	1.04 (0.86, 1.26)	0.89 (0.75, 1.06)	1.00 (0.82, 1.23)	1.05 (0.94, 1.18)
Adjusted 2: RR (95% CI)	1.03 (0.70, 1.51)	0.83 (0.55, 1.26)	0.99 (0.68, 1.43)	1.08 (0.91, 1.28)
Crying spells				
Unadjusted RR (95% CI)	1.21 (0.94, 1.56)	1.06 (0.76, 1.48)	1.00 (0.74, 1.35)	0.99 (0.82, 1.19)
Adjusted 1: RR (95% CI)	1.28 (0.73, 2.25)	0.89 (0.55, 1.47)	1.37 (0.76, 2.46)	0.95 (0.78, 1.17)
Adjusted 2: RR (95% CI)	1.28 (0.73, 2.25)	1.05 (0.51, 2.18)	1.37 (0.76, 2.46)	1.08 (0.76, 1.54)

CRP, high sensitivity c-reactive protein; EAGeR, Effects of Aspirin in Gestation and Reproduction; FGF23, fibroblast growth factor 23; iPTH, intact parathyroid hormone; RR, risk ratio. Bold results are significant at $\alpha = 0.05$.

*Indicates comparisons that survived adjustment for false discovery rate.

Adjusted 1 model controls for age, BMI, smoking status, race, income, exercise, and season of blood draw.

Adjusted 2 model controls for the variables in Adjusted 1 as well as depression, education, alcohol, CRP, and parity.

were included to improve generalizability to the broader population of reproductive aged women. However, it is important to acknowledge that though women met certain cycle length criteria to enroll in the study, some women did report some cycle irregularity. The BioCycle Study had limited power due to the sample size given that the substudy was originally powered to assess changes across the menstrual cycle and associations with hormone levels and the cost associated with multiple longitudinal measurements (Harmon *et al.*, 2020). Additionally, 25(OH)D sufficiency was based on cut points from the Endocrine Society which have been established with respect to bone health (Holick *et al.*, 2011), though levels needed for optimal reproductive health endpoints are unknown. We were limited in assessing other cut points due to small numbers of women with levels below 20 or over 40 ng/ml.

In conclusion, we observed associations between insufficient serum 25(OH)D levels and individual physical symptoms, specifically breast fullness/tenderness, and generalized aches and pain. These findings highlight the potential for 25(OH)D to influence specific symptoms among women with regular menstrual cycles. Given these findings, further research on women with irregular cycles and more extreme symptoms is warranted.

Supplementary data

Supplementary data are available at *Human Reproduction* online.

Data availability

The data underlying this article will be shared on reasonable request to the corresponding author.

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Authors' roles

Z.A., D.L.K., R.M.S., and S.L.M. designed research; D.L.K., A.P.S., and S.L.M. analyzed data; Z.A., D.L.K., and J.G.R. wrote the paper; K.Kim, J.G.R., S.F.Y., L.A.S., N.J.P., R.M.S., A.Z.P., M.T., A.P.S., K.K., and S.L.M. carefully reviewed the manuscript and revised for important intellectual content. S.L.M had primary responsibility for final content. All authors read and approved the final manuscript.

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Conflict of interest

The authors have no conflicts of interest to disclose.

References

Bertone-Johnson ER, Chocano-Bedoya PO, Zagarins SE, Micka AE, Ronnenberg AG. Dietary vitamin D intake, 25-hydroxyvitamin D3 levels and premenstrual syndrome in a college-aged population. *J Steroid Biochem Mol Biol* 2010;**121**:434–437. doi:10.1016/j.jsbmb.2010.03.076

Bertone-Johnson ER, Hankinson SE, Bendich A, Johnson SR, Willett WC, Manson JE. Calcium and vitamin D intake and risk of incident premenstrual syndrome. *Arch Intern Med* 2005;**165**:1246–1252. doi:10.1001/archinte.165.11.1246

Bertone-Johnson ER, Hankinson SE, Forger NG, Powers SI, Willett WC, Johnson SR, Manson JE. Plasma 25-hydroxyvitamin D and risk of premenstrual syndrome in a prospective cohort study. *BMC Womens Health* 2014;**14**:56.doi:10.1186/1472-6874-14-56

Borenstein JE, Dean BB, Endicott J, Wong J, Brown C, Dickerson V, Yonkers KA. Health and economic impact of the premenstrual syndrome. *J Reprod Med* 2003;**48**:515–524.

Craig CL, Marshall AL, Sjöström M, Bauman AE, Booth ML, Ainsworth BE, Pratt M, Ekelund U, Yngve A, Sallis JF. *et al.* International physical activity questionnaire: 12-country reliability and validity. *Med Sci Sports Exerc* 2003;**35**:1381–1395. doi:10.1249/01.Mss.0000078924.61453.Fb

Direkvand-Moghadam A, Sayehmiri K, Delpisheh A, Kaikhavandi S. Epidemiology of premenstrual syndrome (PMS) – a systematic review and meta-analysis study. *J Clin Diagn Res* 2014;**8**:106–109. doi:10.7860/jcdr/2014/8024.4021

Greenland S, Pearl J, Robins JM. Causal diagrams for epidemiologic research. *Epidemiology* 1999;**10**:37–48. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/9888278

Halbreich U, Borenstein J, Pearlstein T, Kahn LS. The prevalence, impairment, impact, and burden of premenstrual dysphoric disorder (PMS/PMDD). *Psychoneuroendocrinology* 2003;**28** Suppl 3:1–23.

Harmon QE, Kissell K, Jukic AMZ, Kim K, Sjaarda L, Perkins NJ, Umbach DM, Schisterman EF, Baird DD, Mumford SL. Vitamin D and reproductive hormones across the menstrual cycle. *Hum Reprod* 2020;**35**:413–423. doi:10.1093/humrep/dez283

Hernan MA, Hernandez-Diaz S, Werler MM, Mitchell AA. Causal knowledge as a prerequisite for confounding evaluation: an application to birth defects epidemiology. *Am J Epidemiol* 2002;**155**:176–184. doi:10.1093/aje/155.2.176

Hibler EA, Sardo Molmenti CL, Dai Q, Kohler LN, Warren Anderson S, Jurutka PW, Jacobs ET. Physical activity, sedentary behavior, and vitamin D metabolites. *Bone* 2016;**83**:248–255. doi:10.1016/j.bone.2015.11.016

Hoang MT, Defina LF, Willis BL, Leonard DS, Weiner MF, Brown ES. Association between low serum 25-hydroxyvitamin D and depression in a large sample of healthy adults: the Cooper Center longitudinal study. *Mayo Clin Proc* 2011;**86**:1050–1055. doi:10.4065/mcp.2011.0208

Hofmeister S, Bodden S. Premenstrual syndrome and premenstrual dysphoric disorder. *Am Fam Physician* 2016;**94**:236–240. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/27479626

Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, Murad MH, Weaver CM, Endocrine Society. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab* 2011;**96**:1911–1930. doi:10.1210/jc.2011-0385

Institute of Medicine Committee to Review Dietary Reference Intakes for Vitamin, D., & Calcium. *Dietary reference intakes for calcium and vitamin D*. In: Ross A. C., Taylor C. L., Yaktine A. L., & Del Valle H. B. (eds). The national academies collection: reports funded by National Institutes of Health. Washington (DC): National Academies Press (US) National Academy of Sciences, 2011.

Khan QJ, Kimler BF, Reddy PS, Sharma P, Klemp JR, Fabian CJ. Randomized trial of vitamin D3 to prevent worsening of musculoskeletal symptoms and fatigue in women with breast cancer starting adjuvant letrozole: The VITAL trial. *Jco* 2012;**30**:9000. doi:10.1200/jco.2012.30.15_suppl.9000

Khan QJ, Reddy PS, Kimler BF, Sharma P, Baxa SE, O'Dea AP, Klemp JR, Fabian CJ. Effect of vitamin D supplementation on serum 25-hydroxy vitamin D levels, joint pain, and fatigue in women starting adjuvant letrozole treatment for breast cancer. *Breast Cancer Res Treat* 2010;**119**:111–118. doi:10.1007/s10549-009-0495-x

Krishnan AV, Feldman D. Mechanisms of the anti-cancer and anti-inflammatory actions of vitamin D. *Annu Rev Pharmacol Toxicol* 2011;**51**:311–336. doi:10.1146/annurev-pharmtox-010510-100611

Lagunova Z, Porojnicu AC, Lindberg F, Hexeberg S, Moan J. The dependency of vitamin D status on body mass index, gender, age and season. *Anticancer Res* 2009;**29**:3713–3720. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/19667169

Lasco A, Catalano A, Benvenga S. Improvement of primary dysmenorrhea caused by a single oral dose of vitamin D: results of a randomized, double-blind, placebo-controlled study. *Arch Intern Med* 2012;**172**:366–367. doi:10.1001/archinternmed.2011.715

Lesher LL, Matyas RA, Sjaarda LA, Newman SL, Silver RM, Galai N, Hovey KM, Wactawski-Wende J, Emerick L, Lynch AM. *et al.* Recruitment for longitudinal, randomised pregnancy trials initiated preconception: lessons from the effects of aspirin in gestation and reproduction trial. *Paediatr Perinat Epidemiol* 2015;**29**:162–167. doi:10.1111/ppe.12177

Morales L, Pans S, Verschueren K, Van Calster B, Paridaens R, Westhovens R, Timmerman D, De Smet L, Vergote I, Christiaens M-R. *et al.* Prospective study to assess short-term intra-articular and tenosynovial changes in the aromatase inhibitor-associated arthralgia syndrome. *J Clin Oncol* 2008;**26**:3147–3152. doi:10.1200/jco.2007.15.4005

Noble K, Matthew A, Burdyga T, Wray S. A review of recent insights into the role of the sarcoplasmic reticulum and Ca entry in uterine smooth muscle. *Eur J Obstet Gynecol Reprod Biol* 2009;**144**(Suppl 1):S11–S19. doi:10.1016/j.ejogrb.2009.02.010

Prieto-Alhambra D, Javaid MK, Servitja S, Arden NK, Martinez-García M, Diez-Perez A, Albanell J, Tusquets I, Nogues X. Vitamin D threshold to prevent aromatase inhibitor-induced arthralgia: a prospective cohort study. *Breast Cancer Res Treat* 2011;**125**:869–878. doi:10.1007/s10549-010-1075-9

Quintana-Zinn FA, Whitcomb BW, Ronnenberg AG, Bigelow C, Houghton SC, Bertone-Johnson ER. Premenstrual symptom patterns and behavioral risk factors in young women: a cross-sectional study. *J Womens Health (Larchmt)* 2017;**26**:1099–1105. doi:10.1089/jwh.2016.5921

Rad M, Sabzevary MT, Dehnavi ZM. Factors associated with premenstrual syndrome in female high school students. *J Educ Health Promot* 2018;**7**:64.doi:10.4103/jehp.jehp_126_17

Radloff LS. The CES-D Scale: a self-report depression scale for research in the general population. *Appl Psychol Meas* 1977;**1**:385–401.

Rastelli AL, Taylor ME, Gao F, Armamento-Villareal R, Jamalabadi-Majidi S, Napoli N, Ellis MJ. Vitamin D and aromatase inhibitor-induced musculoskeletal symptoms (AIMSS): a phase II, double-blind, placebo-controlled, randomized trial. *Breast Cancer Res Treat* 2011;**129**:107–116. doi:10.1007/s10549-011-1644-6

Schafer AS, Shoback DM. *Hypocalcemia: Diagnosis and treatment.* In: De Groot LJ, Chrousos G, Dungan K (eds), Endotext [Internet]. South Dartmouth, MA: MDText.com, 2016.

Schisterman EF, Silver RM, Lesher LL, Faraggi D, Wactawski-Wende J, Townsend JM, Lynch AM, Perkins NJ, Mumford SL, Galai N. Preconception low-dose aspirin and pregnancy outcomes: results from the EAGeR randomised trial. *Lancet* 2014;**384**:29–36. doi:10.1016/S0140-6736(14)60157-4

Schisterman EF, Silver RM, Perkins NJ, Mumford SL, Whitcomb BW, Stanford JB, Lesher LL, Faraggi D, Wactawski-Wende J, Browne RW. *et al.* A randomised trial to evaluate the effects of low-dose aspirin in gestation and reproduction: design and baseline characteristics. *Paediatr Perinat Epidemiol* 2013;**27**:598–609. doi:10.1111/ppe.12088

Shapiro AC, Adlis SA, Robien K, Kirstein MN, Liang S, Richter SA, Lerner RE. Randomized, blinded trial of vitamin D3 for treating aromatase inhibitor-associated musculoskeletal symptoms (AIMSS). *Breast Cancer Res Treat* 2016;**155**:501–512. doi:10.1007/s10549-016-3710-6

Singer O, Cigler T, Moore AB, Levine AB, Do HT, Mandl LA. Hypovitaminosis D is a predictor of aromatase inhibitor musculoskeletal symptoms. *Breast J* 2014;**20**:174–179. doi:10.1111/tbj.12227

Sjaarda LA, Radoc JG, Flannagan KS, Mumford SL, Kim K, Perkins NJ, Silver RM, Schisterman EF. Urinary selective serotonin reuptake inhibitors across critical windows of pregnancy establishment: a prospective cohort study of fecundability and pregnancy loss. *Fertil Steril* 2020;**114**:1278–1287. doi:10.1016/j.fertnstert.2020.06.037

Sternfeld B, Swindle R, Chawla A, Long S, Kennedy S. Severity of premenstrual symptoms in a health maintenance organization population. *Obstet Gynecol* 2002;**99**:1014–1024.

Tague SE, Smith PG. Vitamin D receptor and enzyme expression in dorsal root ganglia of adult female rats: modulation by ovarian hormones. *J Chem Neuroanat* 2011;**41**:1–12. doi:10.1016/j.jchemneu.2010.10.001

Takeuchi A, Okano T, Ishida Y, Kobayashi T. Effects of dietary vitamin D intake on plasma levels of parathyroid hormone and vitamin D metabolites in healthy Japanese. *Miner Electrolyte Metab* 1995;**21**:217–222.

Thomas MK, Lloyd-Jones DM, Thadhani RI, Shaw AC, Deraska DJ, Kitch BT, Vamvakas EC, Dick IM, Prince RL, Finkelstein JS. Hypovitaminosis D in medical inpatients. *N Engl J Med* 1998;**338**:777–783. doi:10.1056/nejm199803193381201

Thys-Jacobs S. Micronutrients and the premenstrual syndrome: the case for calcium. *J Am Coll Nutr* 2000;**19**:220–227.

Wactawski-Wende J, Schisterman EF, Hovey KM, Howards PP, Browne RW, Hediger M, Liu A, Trevisan M, BioCycle Study Group. BioCycle study: design of the longitudinal study of the oxidative stress and hormone variation during the menstrual cycle. *Paediatr Perinat Epidemiol* 2009;**23**:171–184. doi:10.1111/j.1365-3016.2008.00985.x

Waltman NL, Ott CD, Twiss JJ, Gross GJ, Lindsey AM. Vitamin D insufficiency and musculoskeletal symptoms in breast cancer survivors on aromatase inhibitor therapy. *Cancer Nursing* 2009;**32**:143–150. doi:10.1097/01.ncc.0000339262.44560.92

Yamamoto K, Okazaki A, Sakamoto Y, Funatsu M. The relationship between premenstrual symptoms, menstrual pain, irregular menstrual cycles, and psychosocial stress among Japanese college students. *J Physiol Anthropol* 2009;**28**:129–136. doi:10.2114/jpa2.28.129