Obesity and Menstrual Irregularity: Associations With SHBG, Testosterone, and Insulin

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This cross-sectional study aimed to examine the association between different body composition measures, menstrual cycle characteristics, and hormonal factors in a population-based sample of young women. The study sample included 726 Australian women aged 26–36 years who were not currently taking hormonal contraceptives and were not currently pregnant or breast feeding. Anthropometric measures included BMI, waist circumference (WC), and waist–hip ratio (WHR). Menstrual cycle characteristics were self-reported and usual cycles defined as short (≤25 days), normal (26–34 days), or long (≥35 days). Cycles were defined as irregular if there were ≥15 days between the longest and shortest cycle in the past 12 months. Fasting serum levels of sex hormone-binding globulin (SHBG), testosterone, insulin, and glucose were measured and the free androgen index (FAI) derived. Compared with those of normal weight, obese women had at least a twofold greater odds of having an irregular cycle, whether defined by BMI (odds ratio (OR) = 2.61; 95% CI = 1.28–5.35), WC (OR 2.28; 95% CI = 1.16–4.49), or WHR (OR = 2.27; 95% CI = 1.09–4.72). Body composition measures were significantly positively associated with fasting insulin, testosterone, and FAI, and negatively associated with SHBG ($P < 0.01$). Fasting insulin, SHBG, and FAI had the strongest influence on the associations between obesity and irregular cycles, with statistically significant ORs of having an irregular cycle being attenuated to near null values following adjustment. In conclusion, both overall and central obesity were significantly associated with having an irregular menstrual cycle. This association was substantially influenced by hormonal factors, particularly insulin and SHBG.

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INTRODUCTION

Several studies have shown that obese women are more likely to experience menstrual cycle irregularity than non-obese women (1–6). Many of these studies, however, have been limited by small sample sizes and the selection of study participants from patients with gynecological problems or infertility, or from women participating in weight reduction programs (3–6). Some studies have relied on self-reported height and weight (3,5,6) and the definitions of menstrual irregularity have varied widely. Given the rising prevalence of overweight and obesity, it is important to investigate their effects on women's reproductive health and to better quantify the strength of association with menstrual irregularity in population-based samples.

It has been suggested that centrally distributed body fat may be more strongly associated with menstrual abnormalities and adverse hormonal profiles than measures of peripheral body fat or overall adiposity such as BMI (4,5,7). A study of 11,791 20- to 39-year-old members of a weight control organization (5) showed that the risk of oligomenorrhea (cycles >36 days in length) was significantly higher (RR = 3.15, $P < 0.001$) in [Q2] women with upper body fat predominance defined by waist–hip ratio (WHR) >0.80 compared with women with lower body fat predominance (WHR <0.73). A study of 83 obese gynecology patients aged 20–39 years (4) with body fat measured by dual-energy X-ray absorptiometry, found that trunk–leg fat ratio was significantly higher in obese women with oligomenorrhea or secondary amenhorrea than in those with regular menstruation ($P < 0.01$), but overall adiposity did not differ between the two groups.

Increased testosterone and decreased sex hormone-binding globulin (SHBG) have been associated with obesity both in central and peripheral adiposity (8–10). A population-based study of 611 women aged 25–50 years explored the associations between testosterone concentrations and body composition (11) and found a positive dose–response relationship. Higher levels of testosterone have been associated with polycystic ovary syndrome (10) which is related to ovulatory dysfunction

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and menstrual irregularity. Clinical studies have also shown elevated total and free androgen levels and depressed SHBG in obese women with amenorrhea or oligomenorrhea (12,13). These results suggest that testosterone and SHBG may play an important role in the development of menstrual irregularity in obese women.

The purpose of this study was to examine the association between different measures of body composition and menstrual cycle characteristics in a population-based sample of young Australian women. In addition, we sought to evaluate the potential mediating role of serum levels of total testosterone, SHBG, the free androgen index (FAI), and fasting insulin and glucose.

METHODS AND PROCEDURES

Study participants

This study utilized data from the Childhood Determinants of Adult Health study, a 20-year follow-up of children who participated in the 1985 Australian Schools Health and Fitness Survey at age 7–15 years. Details of the 1985 sampling strategy have been described elsewhere (14). At follow-up 6,840 of the original 8,498 study participants (80%) were successfully traced; of these 5,170 agreed to participate in the follow-up study and completed a short enrolment questionnaire. In 2004–2006, a total of 2,410 participants (48% men, 52% women) completed additional questionnaires and attended one of 34 study clinics for extensive physical measurements.

Eligible participants for this study were women who attended study clinics, had completed study questionnaires, were not currently taking hormonal contraceptives (including oral contraceptives, contraceptives implanted subdermally, and progestin releasing intrauterine devices), had not had a hysterectomy, and were not currently pregnant or breast feeding. Of the 1,260 women who completed the questionnaire and attended a study clinic, 447 were current hormonal contraceptive users, 82 women were pregnant, and 5 women had had a hysterectomy. Following these exclusions, a total of 726 participants remained eligible for this study. Participants provided written informed consent and the study was approved by the Southern Tasmania Health and Medical Human Research Ethics Committee.

Assessment of menstrual cycle characteristics

Menstrual cycle characteristics were obtained by written questionnaire. We defined the menstrual cycle as the time from the first day of one period to the first day of the next. The following questions were then asked: "How long is your usual menstrual cycle?"; "What is the longest cycle you have had in the last 12 months?"; "What is the shortest cycle you have had in the last 12 months?" Based upon the reported number of days in a usual menstrual cycle, we defined participants as having a short cycle (≤25 days), normal cycle (26–34 days), or long cycle (≥35 days). These definitions of short and long cycles, although not typical of those used clinically to define polymenorrhea and oligomenorrhea, are similar to those reported in some previous epidemiological studies (2,15–17) and divided study participants into groups of sufficient size for analysis. Menstrual cycle difference (MCD) was calculated as the difference (in days) between the longest and shortest menstrual cycle in the past 12 months and was classified based on the following cutpoints: ≤7 days, 8–14 days, and ≥15 days. An irregular menstrual cycle was defined as an MCD ≥15 days (18). Some women appeared to have confused usual menstrual cycle length and the days of bleeding. After carefully examining individual questionnaire responses, we defined as implausible any responses where usual menstrual cycle was ≤9 days, the longest cycle ≤12 days, or the shortest cycle <8 days. Given the age range of this young adult sample, some women would have likely been pregnant or breast feeding during the previous 12 months; this may have influenced their menstrual cycles and resulted in the extreme values of longest cycles from 174 to 365 days found for five women. They were also treated as implausible values. All implausible values were set as missing in the analysis.

Assessment of body composition

Anthropometric measures were taken at study clinics by trained personnel and taken three times unless the first two measurements were identical. Height was measured without shoes to the nearest 0.1 cm using a portable stadiometer. Weight was measured in light clothing to the nearest 0.1 kg using a digital Heine portable scale. Waist circumference (WC) was measured to the nearest 0.1 cm using a nonstretch steel measuring tape at the narrowest point between the lower costal border and the iliac crest. Hip circumference was measured at the level of the greatest posterior protuberance of the buttocks.

BMI was calculated as the ratio of weight (kg) to height (m) squared (kg/m^2) and was categorized into four groups (<20, 20–24.9, 25–29.9, and ≥30). Obesity was defined as BMI ≥30 (ref. 19). WC was categorized as: <70, 70–79.9, 80–87.9, and ≥88 cm. WHR was calculated by dividing waist by hip circumference and classified as: <0.72, 0.72–0.78, and ≥0.79. Central obesity was defined as WC ≥88 cm or WHR ≥0.79. The rationale for setting the lowest cutpoint of BMI and WC as <20 kg/m^2 and <70 cm, respectively, rather than the established cutpoints of 18.5 kg/m^2 and 80 cm, was based on the observation of "J" shaped curves when the functional relationship between body composition and irregular cycles was checked using fractional polynomials (see Results section for detail).

Hormone measurements

Blood samples were collected after an overnight fast. Fasting glucose was measured enzymatically using the Olympus AU5400 automated analyzer. Plasma insulin was initially measured by a microparticle enzyme immunoassay kit (AxSYM; Abbot Laboratories, Abbot Park, IL) and later, following a change in the choice of kit by the testing laboratory, by electrochemiluminescence immunoassay (Elecsys Modular Analytics E170; Roche Diagnostics, Mannheim, Switzerland). Due to this change in assay methodology, insulin levels from participants' samples (N = 259) assayed using the first methodology were corrected to levels in samples assayed using the second methodology (as per correction factor equation of the laboratory).

Total testosterone concentrations were estimated by radioimmunoassay developed by Repromed Laboratory (Dulwich, South Australia), which is ultrasensitive for lower levels of testosterone (lower limit of sensitivity 347 pmol/l). SHBG was measured using a noncompetitive liquid-phase immunoradiometric assay (SHBG-IRMA kit; Orion Diagnostica, Espoo, Finland). For testosterone, the intra-assay and inter-patient coefficients of variations were 6 and 15% at the 1 nmol/l level, respectively. For SHBG, the interassay and intrapatient coefficients of variations were 2.0–8.6 and 15.4%, respectively. FAI was calculated as: testosterone (nmol/l) × 100/SHBG (units).

Statistical analysis

The main outcome variables were usual menstrual cycle length (short, long) and cycle regularity as quantified by the MCD. Fractional polynomial methods were used to assess the functional shape of the relationship between continuous measures of body composition and each menstrual cycle outcome. Smoothed scatterplots were then used to visually examine these associations and evaluate the appropriateness of *a priori* body composition cutpoints.

Multinomial logistic regression was used to calculate odds ratios (ORs) and 95% confidence intervals (95% CI) for associations between body composition variables and (i) short and long cycle length (normal cycle length as the reference group) and (ii) MCD 8–14 days and MCD ≥15 days (MCD ≤7 days as reference group). All ORs were adjusted for age, level of education, age at menarche, marital status, numbers of live births, and current smoking because of the influences of these factors on both body composition measurements and menstrual cycle outcomes. Spearman correlation coefficients were used to estimate crude associations between body composition and testosterone, SHBG, FAI, fasting glucose and insulin. Multinomial logistic regression was also used to examine

the association between hormonal variables and menstrual cycle characteristics. Hormonal values were logarithmically transformed, and then converted to standard units with mean of zero and standard deviation of one to facilitate comparisons.

To assess the extent to which specific hormonal factors may account for the association between obesity and menstrual irregularity, we adjusted for each hormonal factor separately and assessed the change in ORs and 95% CI compared with a baseline model adjusted only for confounders. [Q3] All statistical analyses were performed using Stata version 9.2 (Statacorp, College Station, TX).

RESULTS

Characteristics of study participants

Characteristics of study participants are shown in **Table 1**. Most women (71.4%) had postschool qualifications, were married or living as married (62.6%), and were nonsmokers (78.1%). Approximately 40% of women were either overweight or obese. The prevalence of obesity defined by BMI, WC, and WHR was 16.1, 20.6, and 23.8%, respectively. The median of BMI, WC, and WHR was 23.7 kg/m^2, 76.1 cm, and 0.75, respectively. The median age at menarche was 13.0 years and 54.6% of women had had one or more live births.

Missing data were as follows: 79 women did not report their usual menstrual cycle length and an additional 29 women provided implausible values; 127 women did not provide information on their longest or shortest cycles in the past 12 months and an additional 85 women provided implausible values. This left 618 women with data on usual cycle length and 514 women with data on their longest and shortest menstrual cycles. The median cycle lengths for those defined as having short, long, and irregular cycles were 24, 38, and 30 days, respectively.

Table 1 Characteristics of study participants (*n* = 726)

Characteristics	Number	Percentage	Mean	s.d.
Age (years)	726		31.2	2.6
BMI (kg/m^2)			25.2	5.6
<20	81	11.2		
20–24.9	356	49.2		
25–29.9	170	23.5		
≥30	117	16.1		
WC (cm)			79.0	12.4
<70	173	23.9		
70–79.9	285	39.4		
80–87.9	117	16.1		
≥88	149	20.6		
WHR			0.75	0.06
<0.72	208	28.7		
0.72–0.789	344	47.5		
≥0.79	172	23.8		
Usual menstrual cycle length (days)[a]			29.4	4.9
≤25 (short cycle)	49	7.9		
26–34 (normal cycle)	521	84.3		
≥35 (long cycle)	48	7.8		
MCD (days)[b]			9.5	13.8
≤7	339	66.0		
8–14	88	17.1		
≥15 (irregular cycle)	87	16.9		
Age at menarche (years)			13.0	1.4
Testosterone (nmol/l)			1.6	0.6
SHBG (nmol/l)			51.2	25.3
FAI			4.1	3.5
Insulin (mU/l)			6.7	4.6
Glucose (mmol/l)			4.9	0.4

FAI, free androgen index; MCD, menstrual cycle difference; SHBG, sex hormone-binding globulin; WC, waist circumference; WHR, waist–hip ratio.
Missing values from 2–55 except where indicated: [a]Missing values = 108. [b]Missing values = 212.

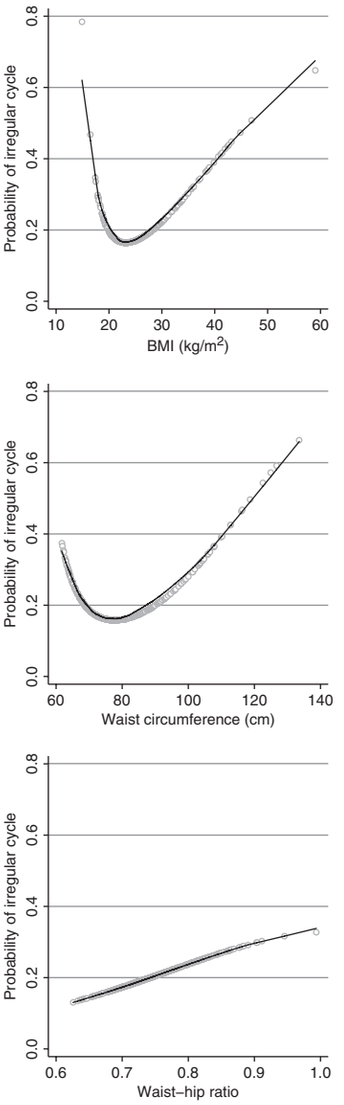

Figure 1 The relationship between the probability of having irregular cycles and body composition measures.

[Q4] **Table 2 Adjusted ORs for various menstrual cycle characteristics across body composition categories**

Body composition	Short cycle (≤25 days)			Long cycle (≥35 days)			MCD 8–14 days			MCD ≥15 days		
	N (%)	OR	95% CI	*N* (%)	OR	95% CI	*N* (%)	OR	95% CI	*N* (%)	OR	95% CI
BMI												
<20	8 (11.8)	1.44	0.54–3.84	8 (11.8)	1.39	0.57–3.40	10 (19.6)	1.59	0.70–3.61	13 (25.5)	2.16	0.96–4.85
20–24.9	22 (7.1)	Ref	Ref	25 (8.1)	Ref	Ref	45 (17.0)	Ref	Ref	37 (14.0)	Ref	Ref
25–29.9	14 (9.5)	1.57	0.74–3.33	6 (4.1)	0.52	0.20–1.37	15 (12.6)	0.76	0.39–1.47	16 (13.4)	0.99	0.51–1.96
≥30	5 (5.5)	0.81	0.28–2.31	9 (9.9)	1.66	0.70–4.00	18 (23.4)	**2.03**	**1.02–4.03**	20 (26.0)	**2.61**	**1.28–5.35**
WC												
<70	13 (9.0)	0.77	0.34–1.72	14 (9.7)	1.09	0.49–2.40	21 (16.7)	0.92	0.49–1.72	24 (19.0)	1.14	0.60–2.17
70–79.9	23 (9.1)	Ref	Ref	17 (6.7)	Ref	Ref	37 (17.6)	Ref	Ref	30 (14.3)	Ref	Ref
80–87.9	8 (7.9)	0.88	0.37–2.10	4 (4.0)	0.54	0.17–1.70	9 (11.7)	0.60	0.27–1.35	8 (10.4)	0.65	0.27–1.54
≥88	5 (4.2)	0.47	0.17–1.31	13 (11.0)	2.22	0.97–5.08	21 (21.2)	1.57	0.81–3.04	25 (25.2)	**2.28**	**1.16–4.49**
WHR												
<0.72	18 (10.1)	Ref	Ref	11 (6.2)	Ref	Ref	29 (18.0)	Ref	Ref	22 (13.7)	Ref	Ref
0.72–0.789	26 (8.7)	0.96	0.48–1.94	23 (7.7)	1.80	0.82–3.96	33 (13.9)	0.96	0.53–1.72	43 (18.1)	1.76	0.95–3.24
≥0.79	5 (3.6)	0.41	0.14–1.20	14 (10.0)	**2.56**	**1.06–6.19**	26 (22.8)	1.83	0.95–3.52	22 (19.3)	**2.27**	**1.09–4.72**

ORs calculated using multinomial logistic regression with a usual menstrual cycle of 26–34 days or a MCD of ≤7 days as the comparison group.
ORs adjusted for age, education, age at menarche, marital status, number of live births, and smoking.
MCD calculated as the difference (days) between the longest and shortest menstrual cycle in the past 12 months.
Boldface values denote statistically significant association.
CI, confidence interval; MCD, menstrual cycle difference; OR, odds ratio; WC, waist circumference; WHR, waist–hip ratio.

Compared with women who provided information on their menstrual cycle characteristics, women with missing values were less well educated (38.4% vs. 28.6% school only) and more likely to be current smokers (35.4% vs. 21.9%) and obese (25% vs. 16.1% BMI ≥30).

Body composition and menstrual cycle characteristics

The functional relationships between continuous measures of body composition and the probability of having an irregular cycle are shown in **Figure 1**. The probability of having an irregular cycle followed an approximate "J" shaped curve across the range of both BMI and WC values, with the prevalence of irregular cycles being elevated at BMI values <20 kg/m^2 and >25 kg/m^2, and at WC values <70 cm and >80 cm. In contrast, the probability of having an irregular cycle increased linearly with increasing values of WHR. Based upon these observations, we utilized the cutpoint of BMI <20kg/m^2 and WC <70 cm, as the threshold for the lower category of BMI and WC in our analysis.

Although there were no significant associations between anthropometric measures and short cycles, these were least common in women with high central adiposity (**Table 2**). In contrast, women with high central adiposity defined by WC (OR = 2.22; 95% CI = 0.97–5.08) or WHR (OR = 2.56; 95% CI = 1.06–6.19) were more likely to have a long cycle compared with their reference groups. Obesity defined by BMI, but not WC or WHR, was significantly associated with the odds of having a variable cycle length (MCD 8–14 days) (OR = 2.03; 95% CI = 1.02–4.03). Compared with those of normal weight, obese women had at least a twofold greater odds of having an irregular cycle (MCD ≥15 days), whether defined by BMI (OR = 2.61; 95% CI = 1.28–5.35), WC (OR = 2.28; 95% CI = 1.16–4.49), or WHR (OR = 2.27; 95% CI = 1.09–4.72).

Table 3 Spearman correlation coefficients for associations of body composition with testosterone, SHBG, FAI, and insulin

Body composition	SHBG	Testosterone	FAI	Insulin
BMI	–0.44	0.17	0.42	0.54
WC	–0.46	0.18	0.43	0.55
WHR	–0.37	0.12*	0.32	0.36

All hormonal variables converted to standard units with mean of 0 and standard deviation of 1.
FAI, free androgen index; SHBG, sex hormone-binding globulin; WC, waist circumference; WHR, waist–hip ratio.
$P < 0.001$ except *$P < 0.01$.

When an alternative common definition of irregular cycles was employed that included both polymenorrhea (usual cycle <21 days) and oligomenorrhea (usual cycles >35 days), associations with obesity were similar for BMI (adjusted OR 2.57; 95% CI = 0.91, 7.22) and somewhat stronger for WC (adjusted OR 2.75; 95% CI = 1.00, 7.61). Similar associations were found between body composition and irregular cycles (MCD ≥15 days) when using the established threshold of underweight as BMI <18.5 (adjusted OR = 2.34, 95% CI = 1.14–4.81) and WC <66 cm (adjusted OR 2.42; 95% CI = 1.25, 4.68).

To examine the possible effect of response bias we analyzed our model using weights based on the inverse probability of response. Weights were formed by constructing a logistic regression model with response/nonresponse as the outcome and the factors associated with nonresponse as covariates (obesity, education, and smoking) (20). Comparing the weighted and unweighted models, we found that the association between

obesity (BMI ≥30) and menstrual irregularity was not changed with weighting (adjusted OR 2.61; 95% CI = 1.28, 5.35 vs. OR 2.61; 95% CI = 1.29, 5.24). Weighting nonresponding types of subjects more heavily did not alter the associations.

The role of sex hormones and insulin

Each of the body composition measures was statistically significantly associated with SHBG, total testosterone concentration, FAI, and fasting insulin (**Table 3**). Hormonal factors were also associated with several menstrual cycle characteristics (**Table 4**). Specifically, higher levels of testosterone and FAI, and lower levels of SHBG, were associated with increased odds of having long and irregular menstrual cycles. Due to an observed U-shaped association between continuous insulin values and the prevalence of irregular cycles, insulin values were categorized at 1 s.d. below and above the mean. Compared with women with insulin values within 1 s.d. of the mean, women with values >1 s.d. above the mean had an increased odds of having irregular cycles (OR = 2.86, 95% CI = 1.51–5.42). Fasting glucose was associated with all body composition measures ($P < 0.01$) but was not associated with any menstrual characteristics ($P > 0.05$)

Because menstrual cycle characteristics and body composition were each associated with SHBG, total testosterone concentration, FAI, and high levels of fasting insulin, we assessed the extent to which specific hormones might account for the association between obesity and menstrual irregularity (**Table 5**). When added individually to a baseline model adjusted for

Table 4 ORs for various menstrual cycle characteristics across standard unit changes in hormonal measures

	Short cycle	Long cycle	MCD 8–14	MCD ≥15 days
	OR (95% CI)	OR (95% CI)	OR (95% CI)	OR (95% CI)
SHBG	1.17 (0.84–1.63)	**0.70 (0.50–0.97)**	0.78 (0.60–1.01)	**0.61 (0.47–0.80)**
Testosterone	1.09 (0.79–1.50)	**1.65 (1.15–2.36)**	**1.52 (1.16–2.00)**	**1.38 (1.05–1.81)**
FAI	0.93 (0.67–1.30)	**1.67 (1.20–2.33)**	**1.52 (1.16–1.97)**	**1.74 (1.32–2.28)**
Insulin				
<–1	0.98 (0.37–2.61)	2.01 (0.83–4.87)	**2.16 (1.74–4.36)**	1.55 (0.69–3.47)
–1 to 1	Ref	Ref	Ref	Ref
>1	0.30 (0.07–1.28)	1.77 (0.77–4.07)	1.49 (0.71–3.12)	**2.86 (1.51–5.42)**

Short cycle, usual menstrual cycle ≤25 days; long cycle = usual menstrual cycle ≥35 days. MCD calculated as the difference (days) between the longest and shortest menstrual cycle in the past 12 months. Insulin categorized based on a U-shaped relationship with irregular cycles.All exposure variables converted to standard units with mean of 0 and s.d. of 1. ORs calculated using multinomial logistic regression using women with a menstrual cycle length of 26–34 days or MCD ≤7 days as the comparison group.
[Q5] Boldface values denote statistically significant association.
CI, confidence interval; FAI, free androgen index; MCD, menstrual cycle difference; OR, odds ratio; SHBG, sex hormone-binding globulin.

Table 5 Association between anthropometric measures and irregular cycles (MCD ≥15days) following adjustment for hormonal factors

	Baseline adjusted[a]		Baseline + SHBG		Baseline + testosterone		Baseline + FAI		Baseline + insulin	
	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	OR (95% CI)
BMI										
<20	2.16	0.96–4.85	2.14	0.90–5.08	1.95	0.83–4.59	2.08	0.87–4.97	2.08	0.91–4.78
20–24.9	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref
25–29.9	0.99	0.51–1.96	0.68	0.32–1.44	0.91	0.45–1.86	0.68	0.32–1.44	0.87	0.43–1.78
≥30	2.61	1.28–5.35	0.99	0.40–2.43	1.87	0.84–4.17	0.98	0.40–2.40	1.11	0.43–2.90
WC										
<70	1.14	0.60–2.17	1.36	0.68–2.73	1.15	0.58–2.28	1.36	0.68–2.74	1.27	0.65–2.48
70–79.9	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref
80–87.9	0.65	0.27–1.54	0.43	0.17–1.11	0.58	0.23–1.45	0.45	0.18–1.17	0.54	0.21–1.37
≥88	2.28	1.16–4.49	0.99	0.44–2.24	1.75	0.84–3.64	1.04	0.47–2.31	1.09	0.45–2.64
WHR										
<0.72	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref
0.72–0.789	1.76	0.95–3.24	1.58	0.81–3.10	1.75	0.91–3.39	1.60	0.81–3.13	1.49	0.79–2.78
≥0.79	2.27	1.09–4.72	1.33	0.57–3.10	2.00	0.91–4.39	1.32	0.57–3.07	1.28	0.56–2.91

CI, confidence interval, FAI, free androgen index; MCD, menstrual cycle difference; OR, odds ratio; SHBG, sex hormone-binding globulin; WC, waist circumference; WHR, waist–hip ratio.
[a]ORs calculated using multinomial logistic regression adjusted for age, education, age at menarche, marital status, numbers of live birth, and smoking.

demographic and reproductive characteristics, fasting insulin, SHBG, and FAI had the strongest influence on associations between body composition and irregular cycle length, with statistically significant ORs (2.27–2.61) being attenuated to near null values (0.98–1.33) following adjustment. Adjustment for total testosterone attenuated these associations to a lesser extent. In addition, adjustment for hormonal factors did not appreciably weaken the positive association between low BMI and irregular cycle length.

DISCUSSION

In this population-based sample of young women who were not currently using hormonal contraceptives, 26% of those who were obese (BMI ≥30) had irregular menstrual cycles compared with 14% of those with a BMI in the range 20–24.9. Similar findings were observed when obesity was defined as a WC ≥88 cm. Although a number of mechanisms may be involved in the association between obesity and irregular menstrual cycles, the finding that the association was substantially attenuated after adjusting for insulin, SHBG, or FAI, suggests that these hormonal factors play an important role. Because the FAI is a measure of testosterone adjusted for SHBG and testosterone alone had a weaker effect, it appears that SHBG is a more important factor than free testosterone. The finding that both insulin and SHBG appear to be on the causal pathway between obesity and menstrual irregularity is consistent with the literature suggesting that increased levels of insulin lead to decreased levels of SHBG (21); though we note recent debate about the relative roles of insulin and glucose in determining SHBG levels in transgenic mice (22)

A strength of the current study was the assessment of both overall and regional body composition. There has been little agreement about whether menstrual irregularity is more strongly associated with overall or central obesity (1,4,5). The probability of having an irregular cycle followed a "J" shaped curve and women with BMI values <20 or >30 kg/m^2 had at least twice the odds of having an irregular cycle compared to normal weight women. The similar "J" shaped relationship of having an irregular cycle length was also found with WC. In contrast, the probability of having an irregular cycle increased linearly with increasing WHR. A previous study (3) also found a "J" shaped curve between BMI and irregular cycles and reported that the lowest prevalence of irregular cycles occurred at BMI values between 20and 22 kg/m^2. Variable cycles (MCD 8–14 days) were also associated with body composition measures but associations were weaker than for irregular cycles (MCD ≥15 days) and only statistically significant for BMI. These findings are in general agreement with prior research suggesting that a low or higher level of adiposity is unfavorable for women's menstrual patterns (1,2,4,6,18,23).

An additional strength of the current study, unique in the literature, was an examination of the potential roles played by testosterone, FAI, SHBG, and insulin levels in the association between body composition and menstrual cycle function in a population-based sample. Both body composition and menstrual irregularity were positively associated with testosterone, FAI, and high insulin values and negatively associated with SHBG. The associations between variable cycles and hormonal factors showed similar patterns to those for irregular cycles, with significantly increased ORs for testosterone and FAI. Adjusting for either FAI, SHBG, or insulin essentially eliminated any association between obesity and irregular cycle length, whereas adjustment for total testosterone resulted in a more modest attenuation of these associations. These observations are in general agreement with previous studies showing body composition measures to be positively associated with total testosterone concentrations in a dose–response manner (11); and increased testosterone levels associated with central and peripheral adiposity (9,10). Few studies, however, have examined the association between androgens, SHBG, and menstrual cycle characteristics. A clinical study (12) examined 11 obese women with or without amenorrhea and 5 nonobese normal cycling women, and found that obese women with amenorrhea had markedly reduced SHBG concentrations compared with cycling nonobese women and significantly increased levels of androgens compared with nonobese cycling and obese cycling women. Even in nonobese healthy women, menstrual irregularities appeared to be associated with higher levels of circulating androgens (24).

This study has several limitations which should be considered when interpreting these findings. The study relied upon written questionnaires for information on menstrual cycle characteristics, rather than menstrual diaries. Previous studies have suggested that women's retrospective self-reports of menstrual cycle lengths can be prone to error (17) and the agreement between diary records and retrospectively recalled menstrual cycle length was moderate (25). If the accuracy of self-reported menstrual characteristics differed by obesity status, then our effect estimates might have been biased. However, although we cannot rule out the possibility of differential error in the reporting of menstrual cycle length by obesity status, previous studies have shown no evidence of this (17,25). Random measurement error would be expected to bias our effect estimates toward the null. There are a range of definitions of menstrual irregularity in the literature. The definition we chose (MCD of ≥15 days) is not the definition used most commonly in clinical practice but it was chosen to capture cycle variability in our study participants and the estimates of its association with obesity were very similar to those observed when a more conventional definition was used.

A second potential limitation of this study was the exclusion of hormonal contraceptive users. Some women may have been prescribed hormonal contraceptives in response to a history of irregular menstrual cycles. To the extent that this occurred, the prevalence of menstrual irregularity would have been underestimated in this study. However, our data show that current hormonal contraceptive users were no more likely than nonusers to have ever seen a doctor for irregular periods (24.6% vs. 25.0%). Third, in this young population sample, some women may have experienced unusual cycles following unrecognized early pregnancy loss and been misclassified as having irregular cycles. To the extent that obesity might increase the risk

of unrecognized pregnancy loss in young women (26), this may have led us to overestimate the association of obesity with menstrual irregularity. However, our finding that the association was substantially attenuated after adjustment for SHBG and FAI suggests that unrecognized pregnancy loss was not a major source of bias.

A further limitation is that we could not examine the role of other female hormones such as estrogen, follicle stimulating hormone, and luteinizing hormone in mediating or confounding the associations between body composition and menstrual cycle characteristics, nor could we determine whether women with irregular or variable cycles were anovulatory or oligo-ovulatory. Although one previous study reported that blood estrogen levels were usually normal or slightly elevated in both premenopausal and postmenopausal obese women (8), other studies have reported increased estrogen, luteinizing hormone levels, or an increased ratio of androgen to estrogen in obese women (27,28). Testosterone levels show some variability through the menstrual cycle with the lowest levels during the first week (29). It would have been ideal to have collected blood for all female participants at a particular time in the menstrual cycle but this was impractical in this study which did not have reproductive health as its primary focus. Any error resulting from this limitation is likely to have been random. Finally, we cannot be certain of the causal direction of the associations observed due to the study's cross-sectional design. A longitudinal study with repeated measures of body composition, hormone levels, and menstrual characteristics would be desirable in future.

In conclusion, this study provides important evidence of the association between both overall and central obesity and menstrual cycle irregularity in young adult women selected from the general population and suggests that these associations are mediated by changes in insulin and SHBG. The degree to which obesity affects female fertility at the population level remains to be elucidated

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DISCLOSURE

The authors declared no conflict of interest.

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