

Elevated body mass index is associated with lower serum anti-mullerian hormone levels in infertile women with diminished ovarian reserve but not with normal ovarian reserve

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Objective: To investigate the association between elevated body mass index (BMI) and ovarian reserve.

Design: Cross-sectional study.

Setting: Academic institutions.

Patient(s): Two hundred ninety women with infertility. Diminished ovarian reserve (DOR) was defined as day 3 FSH >10 IU/L.

Intervention(s): None.

Main Outcome Measure(s): Random serum antimullerian hormone (AMH) levels and number of oocytes retrieved during assisted reproductive technology cycle.

Result(s): Increasing BMI was associated with lower random serum AMH levels in infertile women with DOR but not in women with normal ovarian reserve (NOR). Among women with DOR, mean random serum AMH levels were 33% lower in overweight and obese women compared with women with normal weight. The same association was not true for women with NOR. Out of 290 women evaluated for infertility, 109 women underwent a controlled ovarian hyperstimulation (COH)–IVF cycle. Women with elevated BMI and DOR had lower number of oocytes retrieved compared with women with normal BMI and DOR (6.4 ± 4.3 vs. 9.4 ± 6), an association that was not observed among women with NOR.

Conclusion(s): Overweight and obese women with DOR as defined by high day 3 serum FSH levels have lower serum AMH levels and number of oocytes retrieved compared with nonobese women with DOR. (Fertil Steril® 2011; ■:■–■. ©2011 by American Society for Reproductive Medicine.)

Key Words: Body mass index, obesity, antimullerian hormone, ovarian reserve

Recent data from The National Health and Nutrition Examination Survey showed that the combined prevalence of overweight and obesity was 64% in 2007–2008 among American women. Although there was no statistically significant increase in the rate of overweight and obesity during the last decade, obesity is still at epidemic proportions in the United States (1). Besides being a risk factor for hypertension, diabetes, cardiovascular disease, stroke, and certain cancers, overweight and obesity are also associated with poor reproductive outcomes in most studies (2–4), though the findings are not universal (5). Overweight and obesity are associated with increased amount of gonadotropins used, decreased number of oocytes retrieved, and increased cycle cancellation rates in in vitro fertilization (IVF) cycles (3).

Diminished ovarian reserve (DOR) is a condition associated with decreased quantity and quality of oocytes in the ovaries and is correlated with poor IVF outcomes (6, 7). Markers of ovarian

reserve, including baseline FSH, E₂, inhibin B, antral follicle count, ovarian volume, and, recently, antimullerian hormone (AMH), have been used to counsel patients regarding their reproductive outcomes (7, 8). However, despite the presence of multiple markers, a uniform clinical definition of DOR does not exist (9). In clinical practice, baseline day 3 serum FSH level has been most commonly used to predict ovarian reserve, and levels >10 IU/L are considered to be consistent with DOR. This same level has also been used by the Society for Assisted Reproductive Technology for statistical purposes (10). However, baseline serum FSH is not the best predictor of ovarian response (11, 12). Among others, AMH is emerging as one of the strongest predictors of the number of oocytes retrieved during an IVF cycle (13, 14), and recent reports indicate that AMH may also independently predict pregnancy outcome (15).

A negative correlation between body mass index (BMI) and serum AMH levels has been shown among late reproductive-age women (16) and young women using oral contraceptive pills (17). However, another study investigating the relationship between BMI and ovarian reserve markers, including AMH, failed to demonstrate a similar association (18). To date, it is unclear whether there is a relationship between BMI and AMH, and possible mechanisms underlying this association have not been elucidated.

To our knowledge, an association between ovarian reserve markers (AMH and the number of oocytes retrieved during an

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IVF cycle), and BMI among infertile women with DOR has not been reported. In the present study, we sought to determine whether increasing BMI would adversely affect serum AMH levels and the oocytes retrieved in infertile women with DOR as diagnosed by elevated baseline FSH levels.

MATERIALS AND METHODS

The medical records of 290 women evaluated for infertility at Montefiore Institute for Reproductive Medicine and Health and Genesis Fertility and Reproductive Medicine Center between August 2007 and March 2010 were reviewed. All patients who had their baseline FSH, BMI, and AMH levels recorded during their evaluation were included in the study, which was approved by the Institutional Review Boards of both institutions.

Women were divided into 2 groups based on their baseline serum FSH levels: Group 1 consisted of women with normal ovarian reserve (NOR) with a baseline FSH level ≤ 10 IU/L; group 2 consisted of women with DOR with a baseline serum FSH level >10 IU/L. FSH was measured on day 2 or 3 of the menstrual cycle by using an automated analyzer (Immulate 2000 FSH, Siemens, Deerfield, IL). The sensitivity of the assay was 0.1 IU/L and intra- and interassay coefficients of variation were 2.9%–4.2% and 4.1%–7.9%, respectively. Random serum AMH levels, unrelated to cycle day, were measured in a commercial laboratory (Reprosource, Woburn, MA), based on research-use-only materials and reagents from Beckman Coulter DSL (Chaska, MN) and applied uniformly for all patient samples. Intra- and interassay coefficients of variation with serum control samples were 5%–9% and 7%–12%, respectively (19). Baseline characteristics of women, including age and BMI, were retrieved from their medical records. Of the 290 women, 109 underwent controlled ovarian hyperstimulation (COH)–IVF/intracytoplasmic sperm injection cycles, and their first cycle was included in the study. Baseline maximum FSH and random serum AMH levels, type of protocol, total amount of gonadotropins used (IU), maximum E_2 , and number of oocytes retrieved were examined for each woman. The type of protocol was determined by the patient's doctor, based on her age and the status of ovarian reserve markers.

Data are presented as mean \pm SD. Spearman rank correlation test was used to test the association between BMI and AMH in NOR and DOR women. Wilcoxon rank sum test was used to compare mean AMH levels and mean number of oocytes retrieved among women with normal and high BMI. Linear regression analysis was used to measure the association between BMI and AMH after controlling for age and between BMI and number of oocytes retrieved after controlling for age and type of protocol used. For linear regression analyses, logarithmic transformation was done for AMH

and BMI, and square root transformation was done for oocytes. Statistical analyses were performed using the Stata/IC 10.1 program, and a P value of $<.05$ was considered to be statistically significant.

RESULTS

Demographic and cycle characteristics of the study population are presented in Table 1. There was no statistically significant age difference between NOR and DOR women. Interestingly, women with DOR had statistically significantly lower BMI than women with NOR. As expected, women with NOR had lower baseline FSH, higher AMH, higher E_2 , and higher number of oocytes retrieved and used less gonadotropin. Women with DOR also used flare protocol more often than women with NOR.

The correlation between BMI and AMH is shown in Figure 1. When all study women were included, there was no correlation between BMI and AMH (Fig. 1A). Similarly, no association was found between BMI and AMH among women with NOR (Fig. 1B), even after controlling for age. However, BMI was inversely correlated with AMH among women with DOR ($r = -0.26$; $P = .001$; Fig. 1C). This association persisted after controlling for age ($P = .01$; $\beta = -0.2$). Among women with NOR, those who were overweight or obese had similar mean AMH levels to those who had normal BMI (2 ± 2.4 vs. 1.8 ± 1.5 , respectively; $P = .3$; Fig. 2A). However, among women with DOR, overweight and obese women had 33% lower AMH levels than those with a normal BMI (0.4 ± 0.3 vs. 0.6 ± 0.5 , respectively; $P = .0001$; Fig. 2B).

One hundred nine women underwent a COH-IVF cycle after their infertility evaluation. There was no statistically significant association between BMI and the number of oocytes retrieved when all women were considered. Similarly, no association was found between BMI and number of oocytes retrieved among 51 women with NOR. Among 58 women with DOR, elevated BMI was negatively correlated with the number of oocytes retrieved, that approached significance ($r = -0.28$; $P = .07$). Among this group of women, BMI was still negatively correlated with the number of oocytes retrieved after controlling for age and the type of protocol used, with a P value approaching significance ($P = .07$; $\beta = -0.3$). Although the mean number of oocytes retrieved was similar among NOR women with normal BMI compared with women with elevated

TABLE 1

Baseline and cycle characteristics of women studied.

	NOR (n = 138)	DOR (n = 152)	P value
Age (y)	37.1 \pm 4.8	38.1 \pm 5.2	.1
FSH (IU/L)	7.3 \pm 1.7	15.1 \pm 5	<.0001
BMI (kg/m ²)	27.3 \pm 5.9	25.3 \pm 5.1	.003
AMH (ng/mL)	2 \pm 2	0.5 \pm 0.4	<.0001
Type of protocol ^a			
Gonadotropin only	6 (12)	4 (7)	.5
Gonadotropin + leuprolide flare	7 (14)	25 (43)	.001
Gonadotropin + agonist suppression	12 (23)	7 (12)	.1
Gonadotropin + antagonist suppression	26 (51)	22 (38)	.2
Total gonadotropin used ^a (IU)	4,402 \pm 2,077	6,496 \pm 3,213	<.0001
Maximum E_2 ^a (pg/mL)	2,378 \pm 1,147	1,897 \pm 1,251	.04
No. of oocytes retrieved ^a	10.5 \pm 4.4	8.2 \pm 5.5	.002

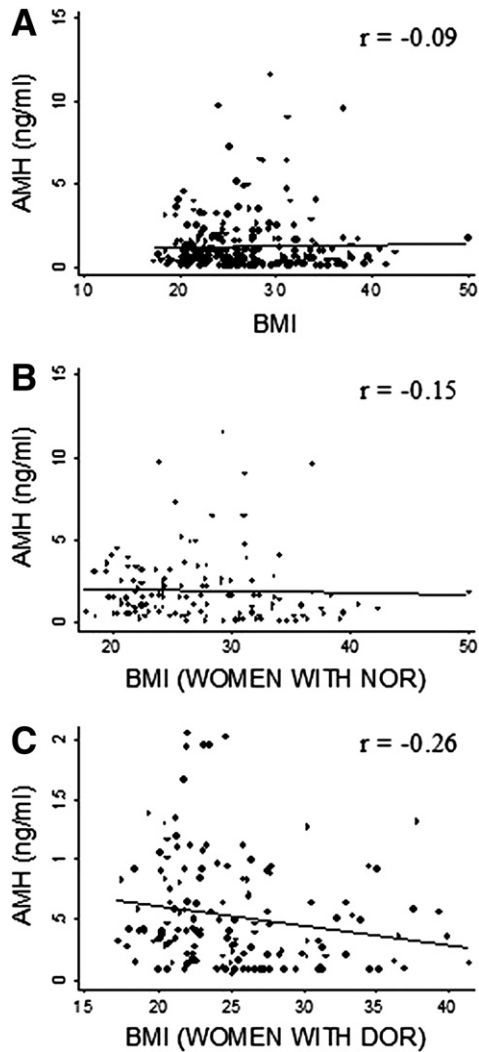
Note: Data presented as mean \pm SD or n (%). AMH = antimüllerian hormone; BMI = body mass index; DOR = diminished ovarian reserve; NOR = normal ovarian reserve.

^a NOR: n = 51; DOR: n = 58.

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FIGURE 1

Increased body mass index (BMI) is associated with decreased antimüllerian hormone (AMH) in women with diminished ovarian reserve (DOR). (A) When all infertile women are included, no association is seen between BMI and AMH ($P = 0.1$). (B) BMI does not correlate with AMH among women with normal ovarian reserve (NOR) ($P = 0.07$). (C) BMI is negatively and statistically significantly associated with AMH among women with DOR ($P = 0.001$).



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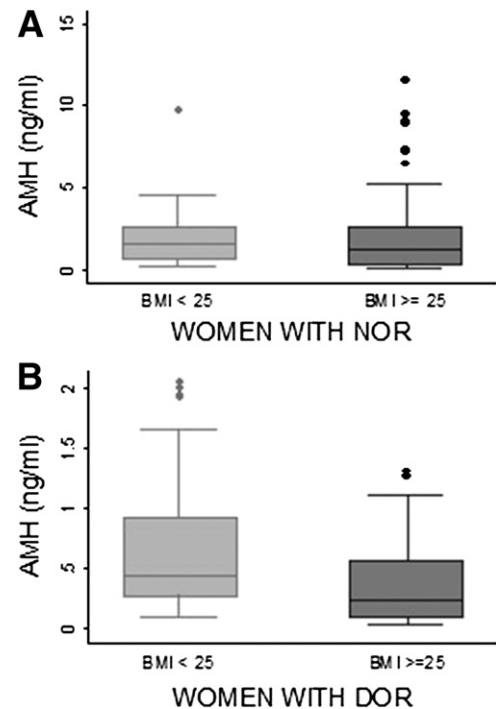
BMI (10.8 ± 4.8 vs. 10.3 ± 4.1 , respectively; $P=.7$), in the DOR group, women with elevated BMI had 32% fewer oocytes retrieved compared with women with normal BMI (6.4 ± 4.3 vs. 9.4 ± 6 , respectively; $P=.03$; Fig. 3A and B).

DISCUSSION

To our knowledge, this is the first study to demonstrate a negative association between BMI, AMH, and the number of oocytes retrieved among infertile women with DOR. A similar negative association had been previously noted among healthy women with late reproductive age (16). In that study, AMH levels were inversely related to BMI and 65% lower in obese women ($BMI \geq 30 \text{ kg/m}^2$) compared

FIGURE 2

BMI affects AMH only in women with DOR. (A) Among women with normal ovarian reserve, mean AMH levels of normal-BMI women are similar to mean AMH levels of women with elevated BMI. (B) Women with elevated BMI in the DOR group have statistically significantly lower mean AMH levels compared with women with normal BMI ($P=.0001$). Abbreviations as in Figure 1.

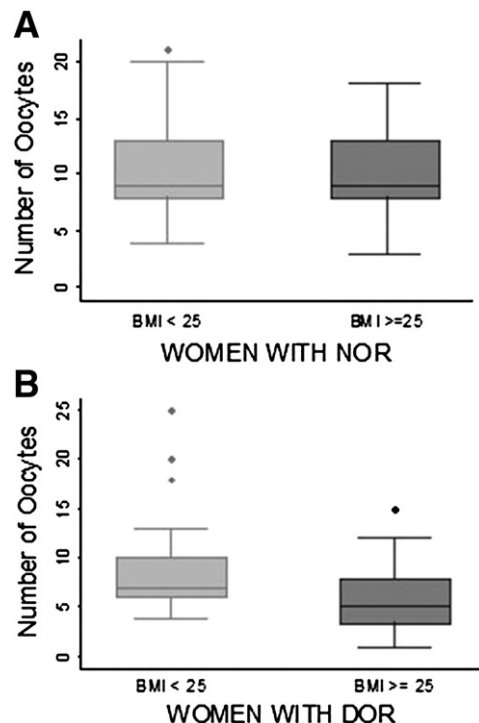


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with nonobese women ($BMI < 30 \text{ kg/m}^2$). Obesity is negatively correlated with other ovarian reserve markers as well, notably inhibin B (20). Overweight and obese premenopausal women have lower inhibin B levels compared with women with normal BMI. The mean age of women in the first study (16) was 45.8 years old. In the second study (20), the ages ranged from 41.4 to 46.7 years old. In contrast to these studies, a recent study that compared ovarian reserve markers, including FSH, AMH, antral follicle count, and ovarian volume, in premenopausal women did not find any difference in these parameters between obese and nonobese women (18). The mean age of women in that population was 46.1 years old, similar to previous studies. However, unlike other studies, they excluded women with a $BMI > 35 \text{ kg/m}^2$. The mean age in our study population was 38 years, which was much younger than the women evaluated in earlier studies. Moreover, our study population was relatively more homogeneous, because it consisted only of women with infertility, and the association was only found in the subgroup of infertile women with DOR. However, despite younger age and differences in the characteristics of the study populations, we found a similar association only in the group of women with DOR who are physiologically more similar to women of late reproductive age in terms of high baseline FSH levels and IVF outcomes. Another study that compared AMH levels in oral contraceptive users found higher AMH levels in obese women ($BMI > 30 \text{ kg/m}^2$) compared with women with normal BMI ($BMI < 25 \text{ kg/m}^2$) (17). Although these were young ovulatory women with a mean age of 29 years, exclusion of

FIGURE 3

BMI affects oocytes only in women with DOR. (A) Among women with normal ovarian reserve, the number of oocytes retrieved from women with normal BMI is similar to the number of oocytes retrieved from overweight or obese women. (B) Overweight or obese women in the DOR group have statistically significantly lower numbers of oocytes retrieved compared with women with normal BMI ($P=.03$). Abbreviations as in Figure 1.



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overweight women (BMI 25–30 kg/m²) and lack of baseline FSH to determine ovarian reserve preclude direct comparison with the present study.

We also found a negative correlation, although statistically nonsignificant ($P=.07$), between BMI and the number of oocytes retrieved among women with DOR, but not among women with NOR. Because AMH is one of the strongest predictors of number of oocytes retrieved during a COH-IVF cycle, this finding is not surprising and is consistent with the correlation of BMI and AMH. Obesity has been associated with poor outcomes in COH-IVF cycles in some (2) but not all (5, 21) studies. Inconsistencies in the definitions of normal BMI as well as overweight and obesity as well as wide variations in the reporting of outcome measures may be responsible for contrasting results. For example, one study reported increased cancellation rates and decreased number of oocytes retrieved

among obese women with NOR, but obesity was defined as BMI >27 kg/m² (22). A meta-analysis of 11 studies that defined BMI according to World Health Organization criteria demonstrated that overweight and obesity are associated with decreased number of oocytes retrieved and decreased pregnancy rates in a population of infertile women (4). Those findings are in accordance with our results, but a subgroup analysis controlling for factors such as age or ovarian reserve was not done. Therefore, it is not possible to attribute the correlation to a particular subgroup of infertile women. Another study that correlated BMI with IVF outcomes reported that increasing BMI has a negative influence on retrieved oocytes, clinical pregnancies, and live births depending on the age of the patient (23). Interestingly, unlike our findings, BMI had a negative influence on IVF outcome in younger women but not older ones, and a recommendation was made to wait for young patients to lose weight before undergoing an IVF cycle. Again, the study did not address the issue of ovarian reserve and is not completely comparable with our data.

The negative correlation of elevated BMI with AMH and number of oocytes retrieved is consistent with previous studies and provides strong evidence that a negative correlation exists between BMI and ovarian reserve, but not in women with NOR. The mechanisms by which obesity may influence ovarian function are not clear. It is unlikely that low serum AMH seen in overweight/obese women with DOR is due to size and dilutional effect, because the same is not true for women with NOR. It is well known that obese women of late reproductive age have more anovulatory and longer cycles compared with nonobese women (24). One of the possible mechanisms is through the interaction of adiponectin with granulosa cells. Adiponectin is secreted from white adipose tissue and its serum levels are decreased in obese women (25). It is present in the follicular fluid of porcine ovaries and its receptors are present on granulosa cells. It induces vascular endothelial growth factor synthesis and modifies enzymes of steroid synthetic pathways, by increasing steroidogenic acute regulatory protein mRNA and decreasing aromatase abundance (26), the same events that take place during the periovulatory process. Therefore, alterations in adiponectin metabolism may lead to dysfunctional ovulatory processes. Consistent with this hypothesis, increased anovulation and decreased in vivo fertilization rates are demonstrated in mice fed a high-fat diet. Moreover, mitochondrial dysfunction and apoptosis rates were markedly increased in the granulosa cells of these mice. Similarly, in the same study, signs of lipotoxicity were observed in the follicular fluid of obese women undergoing COH-IVF compared with normal-weight women (27), which could be a possible explanation for poorer IVF outcomes seen in women with elevated BMI.

In summary, we report a negative correlation between elevated BMI and AMH and oocytes retrieved in infertile women with DOR. The fact that this association is limited to women with DOR, and not observed in women with NOR, may in part explain the discrepancy in the literature regarding the effect of obesity and IVF outcomes. Future clinical and basic studies are needed to elucidate further the complex interactions between obesity and ovarian function.

REFERENCES

1. Flegal KM, Carroll MD, Ogden CL, Curtin LR. Prevalence and trends in obesity among US adults, 1999–2008. *JAMA* 2010;303:235–41.
2. Kupka MS, Gnath C, Buehler K, Dahncke W, Kruessel JS. Impact of female and male obesity on IVF/ICSI: results of 700,000 ART-cycles in Germany. *Gynecol Endocrinol* 2011;27:144–9.
3. Zhang D, Zhu Y, Gao H, Zhou B, Zhang R, Wang T, et al. Overweight and obesity negatively affect the outcomes of ovarian stimulation and in vitro fertilisation: a cohort study of 2628 Chinese women. *Gynecol Endocrinol* 2010;26:325–32.
4. Maheshwari A, Stofberg L, Bhattacharya S. Effect of overweight and obesity on assisted reproductive technology—a systematic review. *Hum Reprod Update* 2007;13:433–44.
5. Dechaud H, Anahory T, Reyffmann L, Loup V, Hamamah S, Hedon B. Obesity does not adversely

- affect results in patients who are undergoing in vitro fertilization and embryo transfer. *Eur J Obstet Gynecol Reprod Biol* 2006;127:88–93.
6. Meden-Vrtovec H. Ovarian aging and infertility. *Clin Exp Obstet Gynecol* 2004;31:5–8.
 7. Derman SG, Seifer DB. In vitro fertilization in the older patient. *Curr Womens Health Rep* 2003;3:375–83.
 8. Seifer DB, MacLaughlin DT, Christian BP, Feng B, Sheldon RM. Early follicular serum mullerian-inhibiting substance levels are associated with ovarian response during assisted reproductive technology cycles. *Fertil Steril* 2002;77:468–71.
 9. Coccia ME, Rizzello F. Ovarian reserve. *Ann N Y Acad Sci* 2008;1127:27–30.
 10. Society for Assisted Reproductive Technology. IVF success rates, national data summary. Available at: <http://www.sart.org/frame/detail.aspx?id=3893>. Accessed December 10, 2010.
 11. Barad DH, Weghofer A, Gleicher N. Comparing anti-Mullerian hormone (AMH) and follicle-stimulating hormone (FSH) as predictors of ovarian function. *Fertil Steril* 2009;91:1553–5.
 12. Nardo LG, Gelbaya TA, Wilkinson H, Roberts SA, Yates A, Pemberton P, et al. Circulating basal anti-mullerian hormone levels as predictor of ovarian response in women undergoing ovarian stimulation for in vitro fertilization. *Fertil Steril* 2009;92:1586–93.
 13. Seifer DB, MacLaughlin DT. Mullerian inhibiting substance is an ovarian growth factor of emerging clinical significance. *Fertil Steril* 2007;88:539–46.
 14. Hazout A, Bouchard P, Seifer DB, Aussage P, Junca AM, Cohen-Bacrie P. Serum antimullerian hormone/mullerian-inhibiting substance appears to be a more discriminatory marker of assisted reproductive technology outcome than follicle-stimulating hormone, inhibin B, or estradiol. *Fertil Steril* 2004;82:1323–9.
 15. La Marca A, Nelson SM, Sighinolfi G, Manno M, Baraldi E, Roli L, Xella S, Marsella T, Tagliasacchi D, D'Amico R, Volpe A. Anti-Müllerian hormone-based prediction model for a live birth in assisted reproduction. *Reprod Biomed Online* 2011;22:341–9.
 16. Freeman EW, Gracia CR, Sammel MD, Lin H, Lim LC, Strauss JF third. Association of antimullerian hormone levels with obesity in late reproductive-age women. *Fertil Steril* 2007;87:101–6.
 17. Steiner AZ, Stanczyk FZ, Patel S, Edelman A. Anti-mullerian hormone and obesity: insights in oral contraceptive users. *Contraception* 2010;81:245–8.
 18. Halawaty S, Elkattan E, Azab H, ElGhamry N, Al-Inany H. Effect of obesity on parameters of ovarian reserve in premenopausal women. *J Obstet Gynaecol Can* 2010;32:687–90.
 19. Seifer DB, Baker VL, Leader B. Age-specific serum anti-Mullerian hormone values for 17,120 women presenting to fertility centers within the United States. *Fertil Steril* 2011;95:747–50.
 20. Gracia CR, Freeman EW, Sammel MD, Lin H, Nelson DB. The relationship between obesity and race on inhibin B during the menopause transition. *Menopause* 2005;12:559–66.
 21. Vilarino FL, Christofolini DM, Rodrigues D, de Souza AM, Christofolini J, Bianco B, et al. Body mass index and fertility: is there a correlation with human reproduction outcomes? *Gynecol Endocrinol*.
 22. Spandorfer SD, Kump L, Goldschlag D, Brodtkin T, Davis OK, Rosenwaks Z. Obesity and in vitro fertilization: negative influences on outcome. *J Reprod Med* 2004;49:973–7.
 23. Sneed ML, Uhler ML, Grotjan HE, Rapisarda JJ, Lederer KJ, Beltsos AN. Body mass index: impact on IVF success appears age-related. *Hum Reprod* 2008;23:1835–9.
 24. Santoro N, Lasley B, McConnell D, Allsworth J, Crawford S, Gold EB, et al. Body size and ethnicity are associated with menstrual cycle alterations in women in the early menopausal transition: The Study of Women's Health Across the Nation (SWAN) daily hormone study. *J Clin Endocrinol Metab* 2004;89:2622–31.
 25. Kadowaki T, Yamauchi T. Adiponectin and adiponectin receptors. *Endocr Rev* 2005;26:439–51.
 26. Ledoux S, Campos DB, Lopes FL, Dobias-Goff M, Palin MF, Murphy BD. Adiponectin induces periovulatory changes in ovarian follicular cells. *Endocrinology* 2006;147:5178–86.
 27. Wu LL, Dunning KR, Yang X, Russell DL, Lane M, Norman RJ, et al. High-fat diet causes lipotoxicity responses in cumulus-oocyte complexes and decreased fertilization rates. *Endocrinology* 2010;151:5438–45.