The Value of Predictive Factors in Optimizing Ovarian Stimulation Treatment Options for Assisted Reproductive Technology

a report by
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Successful outcomes following assisted reproductive technology (ART) are largely dependent on the patient's response to controlled ovarian stimulation (COS), vis à vis the number and quality of oocytes obtained and ultimately the number of embryos available for transfer. Given the marked variability in ovarian response among *in vitro* fertilization (IVF) patients, the choice of stimulation protocol must be individualized, both for women with a history of prior cycles and for first-time IVF patients.

Approximately 4,000–5,000 ovarian stimulation cycles are performed annually at The Center for Reproductive Medicine and Infertility (CRMI), equally divided between IVF and ovulation induction (OI). Patients frequently come to the CRMI following unsuccessful, and often multiple cycles at other clinics in the US and abroad. Approximately half of our patients are older than 38 years and a third are over 40 years.

There are several factors that can predict the ovarian response to COS and, therefore, the likelihood of success following ART (see *Table 1*, page 6), however, there are no absolutes in this regard. The objective of this paper is to review the current understanding of these factors and how they might be used in practice to optimize individual stimulation protocols while minimizing potential complications.

Factors Predictive of Ovarian Response

Age

Age is the most important factor in determining success rates after IVF.[1] As the average age of our patients has increased, the age-related decline in fecundity has increasingly become a dominant cause of subfertility.[2] The loss of fecundity with age appears to be a consequence of both oocyte depletion and reduced oocyte quality. We have previously demonstrated that maternal age can predict over 80% of IVF success.[3]

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The 2004 Centers for Disease Control (CDC) database indicates that per cycle live birth rates vary significantly between age groups (from 43% for women aged <35 to 6% in women >42 years of age).[4] It is of interest to note that many women over the age of 40 continue to pursue fertility treatments utilizing their own oocytes. According to a recent report, in excess of 4,000 IVF cycles in the US were initiated in women over the age of 42.[5] The reduced live birth rate seen in older women is also a result of the increased incidence of miscarriages (pregnancy losses approach 45% by 43 years).[6]

In a review of a large series of consecutive IVF pregnancies (with documented fetal cardiac activity), Spandorfer et al.[5] clearly demonstrated a linear increase in miscarriage rates with advancing maternal age. Eighty-two percent of the pregnancy losses in women aged ≥40 years were associated with chromosomally abnormal fetuses (predominantly trisomies).

The reduced fertility associated with aging is primarily associated with aging of the ovaries and oocytes, rather than of the uterus and endometrium. This was underscored by the observation that donor oocyte cycles resulted in high pregnancy rates among recipient women, irrespective of their age (see *Figure 1*, page 6).[6] However, in a recent large analysis of women receiving donor oocytes, Toner et al.[7] demonstrated that while overall success rates are constant for recipients ranging from their mid-20s through to late 40s, beyond 48 years they decline precipitously.

Assessment of Ovarian Reserve

The only direct method for quantifying follicle numbers and hence ovarian reserve is through an ovarian biopsy.[8] However, the risk–benefit ratio of this procedure does not warrant its use for clinical evaluation of reproductive aging.[9–11]

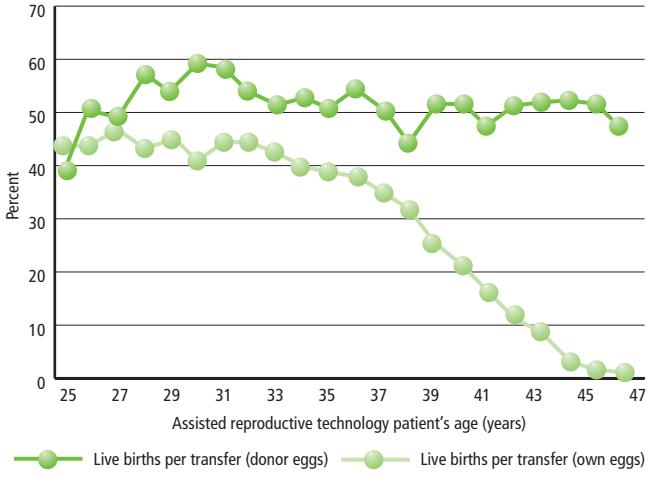
A less invasive method of assessing ovarian reserve is through the sonographic determination of either the ovarian volume or the antral follicle count (see *Figure 2*, page 7). Ovarian volume is estimated using transvaginal ultrasound, with measurements in three planes and application of the formula V = D1 x D2 x D3 x 0.523, with D1, D2, and D3 representing the maximal longitudinal, antero-posterior, and transverse diameters. Mean ovarian volume is the mean volume calculated for both ovaries in an individual patient.

In a study by Lass et al.,[12] the mean ovarian volume in 140 patients varied from 0.5 to 18.9cm^3 and the authors concluded that there was a strong association between ovarian volume and ovarian reserve. In a second study by Syrop et al.,[13] ovarian volumes were determined prior to pituitary down regulation. The median ovarian volume of the smaller of

Table 1: Potential Predictive Factors

Physical and age-related	Chronological age
	Ovarian reserve
	Antral follicle count
	Ovarian volume
	Stromal blood flow
	Menstrual cycle length/regularity
Hormonal	Follicle-stimulating hormone
	Inhibin B
	Anti-Mullerian hormone
	Estradiol
	Luteinizing hormone
Lifestyle	Body-mass index
	Smoking status
	Alcohol consumption
	Caffeine intake

Figure 1: Effect of a Woman's Age on Pregnancy Rate Using Own and Donated Eggs



Permission obtained from Lippincott Williams & Wilkins: Van Voorhis BJ, Outcomes from ART, Obstet Gynecol, *2006;107(1):183–200.*

the two ovaries was 6.4ml with a mean of 7.2±3.6ml; here the 10th percentile was 3.2ml. In a study designed to compare the predictive values of age, basal follicle-stimulating hormone (FSH) level, body-mass index (BMI), total ovarian volume, and total antral follicle count, with respect to the number of oocytes harvested, it was shown that total antral follicle number had the greatest predictive value, followed by basal FSH, BMI, and age.[14] Similarly, in a study of 166 women undergoing IVF, Tomas et al.[15] concluded that the number of small follicles (2–5mm) present before ovarian stimulation was a better predictor of outcome than ovarian volume or age alone. Perhaps there is still a value in measuring ovarian volume in cases where obesity or distant localization of the ovaries diminishes the image quality to such an extent as to preclude accurate follicle counts.

Frattarelli et al.[16] assessed the predictive value of, and defined threshold levels for, basal antral follicle counts (AFC) in 278 patients who had basal ovarian measurements prior to initiating gonadotropin treatment. They found that a basal AFC of <10 or a mean ovarian diameter of <2cm on cycle day three predicted higher cancellation rates and ultimately poor ART outcome. In a recently published meta-analysis including 10 studies of ovarian volume and 17 studies of antral follicle counts, Hendriks et al.[17] concluded that the predictive value of ovarian volume for poor response is clearly inferior compared with that of AFC, and that the AFC should be considered the test of first-choice when estimating quantitative ovarian reserve before IVF.

Hormone Levels

Ovarian reserve is most often measured by assessment of baseline FSH, anti-Mullerian hormone (AMH), inhibin B, and estradiol (E2) levels.

One of the most widely used prognostic tests is the day three basal FSH level. Almost 20 years ago, Muasher et al.[18] studied basal and post-gonadotropin-releasing hormone (GnRH) agonist stimulated serum levels of FSH and luteinizing hormone (LH) on day three of the cycle. We found that basal FSH levels were predictive of estradiol response, oocyte yield, and pregnancy rates. Scott et al.[19] further refined the model and eliminated the assessment of LH after finding that it had little predictive value. Toner et al.[20] concluded that the combination of age and basal FSH is a useful index of functional ovarian reserve and is therefore helpful in counseling patients regarding their prognosis. A recent retrospective analysis of a large cohort of IVF patients undertaken by Roberts et al.[2] indicated that a history of one or more instances of elevated basal FSH levels predicts a poor response to gonadotropins irrespective of age. The authors found that women with a history of three or more elevated FSH levels, regardless of age, did not successfully conceive. Patients aged >40 years with a history of elevated FSH levels manifest both poor ovarian response and compromised embryo quality relative to those with normal FSH levels, and have lower oocyte yield, higher cancellation rates, and lower implantation and pregnancy rates. In patients younger than 40 years, prior elevated basal FSH levels predict a lower oocyte yield even in subsequent IVF cycles when basal FSH levels are normal, but this does not necessarily translate to lower pregnancy or implantation rates.

Basal E2 and inhibin B levels have also been studied as candidate markers for ovarian reserve. E2 was studied extensively in the 1990s, but a systematic review concluded that it is of limited value for both poor response and non-pregnancy.[21] Seifer et al.[22] correlated low day three serum inhibin B levels with reduced response and diminished success rates following ART. In contrast, recent studies[23] suggest that serum inhibin B levels are less predictive of ART outcome than other markers.

More recently, attention has shifted to AMH, which is produced by the granulosa cells of pre-antral and small antral follicles. Van Rooij et al.[24] studied the relationship between AMH levels and ovarian response during ovarian stimulation for IVF in 130 patients undergoing their initial treatment cycle. They found that serum AMH levels were highly correlated with the number of antral follicles and the number of oocytes retrieved. Furthermore, AMH levels were directly correlated with ovarian response. The addition of inhibin B and FSH concentrations to AMH further enhanced the predictive value of the model. Fanchin et al.[25] suggested that AMH may be more robustly correlated with the number of early antral follicles than inhibin B, E2, FSH, and LH on cycle day three. A recent systematic overview[21] concluded that the ovarian reserve tests currently in

use have only modest to poor predictive value. The ideal predictive test is yet to be defined.

Lifestyle

Other extrinsic factors are also worthy of consideration. Obesity, as measured using the BMI, and its secondary effects on insulin resistance and hyperandrogenism, may also affect fertility. Dokras et al. reported a significantly higher risk for IVF cycle cancellation in morbidly obese patients, although BMI had no effect on clinical pregnancy or delivery rate. Furthermore, obese and morbidly obese subjects had a significantly higher risk for obstetric complications.[26]

Cigarette smoking, alcohol, and caffeine are other lifestyle factors that have a negative impact on fertility. Specifically, cigarette smoking is linked to elevated FSH levels, although it does not appear to affect inhibin B and estradiol levels or antral follicle count.[27] There is compelling evidence that smoking has a negative influence on IVF outcome.[28]

Optimizing Treatment With Consideration to Predictive Factors

The highest success rates with IVF or ovulation induction are observed in the first treatment cycle. Patients with the highest reproductive potential conceive in the first one to three cycles. Choice of the appropriate starting protocol is important. It is not always possible to predict response in the treatment-naïve patient and the outcome of the first protocol becomes important in determining how to adjust subsequent treatment regimens. Optimization of the starting dose of gonadotropins FSH is a key component for success.

Normal Responders

Studies of predictive factors are not always comparable owing to differences in study design and subject characteristics. In an attempt to consolidate this information, Popovic-Todorovic et al.[29] examined several of these predictive factors in a single prospective study of 155 women undergoing their first IVF/intracytoplasmic sperm injection (ICSI) treatment cycle. Inclusion criteria were normal basal FSH level, presence of both ovaries, regular menstrual cycle, age less than 40 years, and absence of endocrine disorders—in other words, a relatively 'normal' patient. All patients were started on a standard gonadotropin dose of 150IU/day and measurements were taken of the number of antral follicles, ovarian volume, ovarian stromal blood flow, and hormone levels, including E2, FSH, LH, inhibin B, and testosterone. Age, BMI, cycle length, and smoking status were also recorded. On day eight of stimulation, the response was assessed and the dose of FSH adjusted if necessary.[29] While all the factors measured were, to a certain extent, independent predictors of the number of aspirated follicles, the researchers determined that some were more clinically useful than others. They devised a nomogram consisting of five weighted factors—number of antral follicles, ovarian volume, Doppler score (stromal blood flow), age, and smoking status. According to the nomogram, a 36-year-old non-smoking woman with 10 antral follicles, ovarian volume of 8ml, and a Doppler score of four would receive 220IU/day of FSH; a 28 year-old woman who smokes five cigarettes a day and has 30 antral follicles, ovarian volume of 11ml, and a Doppler score of six would receive 120IU/day of FSH.

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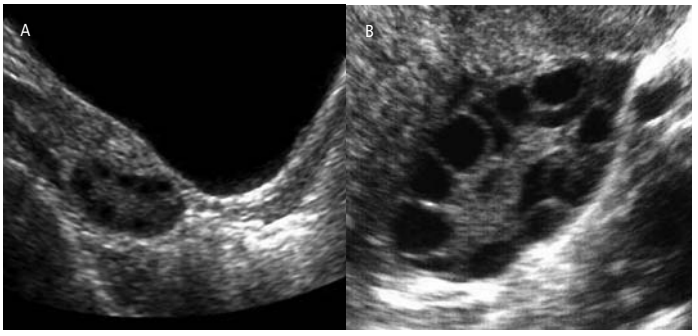
Figure 2: Images of Normal (A) and Polycystic (B) Ovaries



A: Reproduced with permission from Siemens Ultrasound and obgyn.net
B: Reproduced with permission from www.ivf-infertility.com

This nomogram was then compared in a prospective randomized trial with a standardized FSH dose in 267 patients. Retrieval of 5–14 oocytes was deemed an appropriate response. This study suggested that an individualized dosing regimen optimized ovarian response and reduced the need for dose adjustments during COS. A higher ongoing pregnancy rate was observed in the individualized dosing group.[30]

Non-standard Patients—Potential Poor Responders

It is important to distinguish between follicular response to treatment and overall ART success. Older women face a significant impairment in implantation efficiency, which is largely independent of the magnitude of their stimulation response. There are presently no treatment strategies apart from oocyte donation that have been shown to significantly improve implantation efficiency in older women.[3,7] Nonetheless, recent efforts have focused on the continued development of improved stimulation protocols, the facilitation of embryo implantation through *zona pellucida* micromanipulation (assisted hatching), and screening pre-implantation embryos for aneuploidy,[31] among other techniques.[32]

For a subset of women who have failed IVF, the use of autologous endometrial co-culture (AECC) may be of value. This technique improves embryo quality and pregnancy outcome, and Spandorfer et al.[33] showed that histological characteristics, including dating of the endometrium, predict success following AECC. In a prospective study of 86 consecutive patients undergoing IVF, there was a significant improvement in embryo quality using AECC.

Non-standard Patients—Polycystic Ovary Syndrome

Polycystic ovary syndrome (PCOS) is itself a predictor of infertility[34] and is also strongly related to obesity.[35] For PCOS patients in the context of IVF, there is evidence that a protocol combining an oral contraceptive overlapping with a GnRH agonist followed by low-dose

gonadotropins will reduce the risk of ovarian hyperstimulation syndrome (OHSS) while helping to optimize oocyte maturation and pregnancy rates.[36] An alternate approach omits GnRH down regulation while employing a GnRH agonist as the ovulatory stimulus in patients at risk of developing OHSS.[37] Weight and hyperandrogenism are particular concerns for patients with PCOS, which can be addressed individually or in combination (which is discussed at length in a later article by Dokras, see *pages 9–12*).

Conclusion

The greatest predictor of success following fertility treatment is maternal age. Women between the ages of 25 and 30 years have the best prognosis for achieving a live birth, while women over the age of 40 have considerably poorer prospects.

Hormone levels are also useful predictors, particularly FSH, E2, inhibin B, and more recently AMH. FSH is perhaps the most studied so far and is the most widely used. Women with lower basal FSH levels—as measured in the early follicular phase—have the highest chance of pregnancy success.

At the CRMI, we first obtain day three FSH and E2 levels, along with a complete case history and physical exam. In patients who have previously failed IVF, it is critical to examine their response in previous stimulation cycles. We evaluate the antral follicle count by ultrasound and grossly evaluate the ovarian volume. Inhibin B level has been used intermittently, but we do not use it routinely. Based on the previous response, basal hormone testing, age, antral follicle count, and weight, we will recommend a suitable protocol.

Patients at our Center are advised not to smoke, given the potential detrimental effects on steroidogenesis and follicular development. For women with a high BMI, particularly in the setting of PCOS with high basal insulin levels, an insulin-sensitizing agent can be useful, as can a consultation with a nutritionist or a diabetologist to assist with weight loss.

Tailoring the starting dose of gonadotropins in COS to suit individual needs has yielded good results. Algorithms presented in the literature are useful but are infallible. Further well-designed prospective clinical trials will help to refine stimulation protocols and assist in the optimization of treatment. ■

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