The antral follicle count is a better marker than basal follicle-stimulating hormone for the selection of older patients with acceptable pregnancy prospects after in vitro fertilization

This observational study shows that the antral follicle count is a better predictor of ongoing pregnancy in IVF patients aged >38 years of age than is basal FSH. Patients aged <44 years with a normal antral follicle count still have acceptable pregnancy rates after IVF and therefore deserve treatment. (Fertil Steril® 2005;83:811–4. ©2005 by American Society for Reproductive Medicine.)

The ability of a woman to conceive declines with age, both in the natural cycle (1) and after the application of assisted reproductive technologies (ART). Although ART offers the best chances for pregnancy in women of advanced reproductive age (2), the outcome is poor in comparison with younger patients. Nevertheless, some older patients still have good results after IVF treatment, especially those with a normal response to ovarian hyperstimulation (3, 4).

According to national IVF guidelines, treatment of patients aged >40 years is not useful as long as reliable measuring of individual ovarian reserve is not possible (5). Therefore these patients are usually denied IVF treatment in the Netherlands. However, because of the current trend to postpone childbearing, the demand for assisted reproductive technology in older patients is rising. Some of these patients might still have good pregnancy prospects. The question arises as to whether it is possible to select older patients who will still benefit from IVF treatment by using a marker other than chronological age.

The age-related decline in success rates after IVF is assumed to be caused by a diminished ovarian reserve. The basal FSH concentration is widely used as a marker to determine the individual ovarian reserve. However, in two studies comparing predictors of ovarian response in IVF, the antral follicle count (AFC) performed slightly better as a parameter than did basal FSH (6, 7). Older IVF patients generally have fewer antral follicles than younger patients, but a higher AFC is associated with increased pregnancy rates, also in older patients (8).

The aim of the current study was to examine whether the AFC can be used to select older patients with favorable IVF outcome. We also aimed at assessing the performance of the AFC as a predictor of ongoing pregnancy after IVF in older patients in comparison with basal FSH.

Between July 1999 and April 2003, all women aged between 38 and 46 years who started their first IVF or intracytoplasmic sperm injection (ICSI) treatment in our IVF center were prospectively included in this study. It has been well documented that success rates in IVF show a progressive decline between 35 and 40 years and are low after the age of 40 years (9–12). The cutoff of 38 years that we used in the present study was based on a previous study that we conducted in our center. In this study, it was shown that implantation rates decline rapidly after the age of 37 years (13). In our clinic, patients aged ≥41 years are usually not allowed IVF but could enter the program for observational studies if they still had a regular menstrual cycle. There were no restrictions regarding the basal FSH level.

Age was calculated as completed years on the day that the ovarian stimulation was started. The basal FSH values of the patients participating in the present study were retrospectively collected from the clinical chemistry laboratory database. Basal FSH was measured with the automated immunometric FSH assay (Chiron Diagnostics, Tarrytown, NY) on the automated ACS-180 immunoassay platform (Bayer, Tarrytown, NY). All patients underwent an ovarian ultrasonography after ovarian down-regulation (14, 15), just before the start of the stimulation with gonadotropins. A Voluson 530D (Kretz Technik, Zipf, Austria) was used with a 7.5-MHz vaginal transducer. The images were stored, and two observers recorded all antral follicles sized ≤5 mm afterwards.

We have permission of our institutional review board to use anonymous data that are collected during the course of fertility treatment, provided that patients are aware of this and do not object. In all our patient information, this procedure is extensively explained. None of the patients included in this study objected to the use of their data.

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Participation in the study did not involve any deviation from our standard protocol.

The patients were treated with a long suppression protocol. Down-regulation with leuprolide acetate injections (Lucrin; Abbott, Hoofddorp, The Netherlands) was started in the midluteal phase at a dose of 1 mg/d, followed by ovarian stimulation with follitropinα (Gonal-F; Serono Benelux BV, The Hague, The Netherlands). The standard starting dose was 150 IU. In 44 patients, the starting dose was adjusted because of an expected poor or high response, based on our clinical practice. The distribution of these cases between the groups studied (normal basal FSH versus normal AFC and elevated FSH versus low AFC) was comparable. The stimulation was monitored by using ovarian ultrasound and E_2 measurements. When necessary, the dose was adapted after 7 days.

When at least one follicle reached 18 mm, 10,000 IU of hCG (Profasi; Serono Benelux) was administered, and 36 hours later, the transvaginal oocyte collection took place. A maximum of three embryos was transferred. The luteal phase was supplemented with hCG or with micronized P (Progestan; Nourypharma BV, Oss, the Netherlands). The details of the protocol used in our center have been published elsewhere (13).

The primary outcome measure was ongoing pregnancy (fetal heart activity on ultrasound beyond 12 weeks of gestation). Secondary outcome measures were clinical pregnancy (positive pregnancy test 18 days after ovum pickup) and normal ovarian response (at least 4 oocytes at the ovum pickup). Data were analyzed with the Statistics Package for Social Sciences for Windows, version 10.1 (Chicago, IL).

The outcome measures were compared between patients with a normal FSH level (<15 IU/L) and patients with an elevated basal FSH level (≥15 IU/L). The cutoff value of 15 IU/L was chosen because a retrospective study that was performed in our center showed poor pregnancy rates in patients with FSH above this level (16). The outcome measures were also compared between patients with fewer than five antral follicles and with five or more antral follicles on ultrasound. We set the cutoff between four and five antral follicles on the basis of the study by Kupesic et al. (8), in which no pregnancies occurred in patients with fewer than five antral follicles on ultrasound before the start of the IVF stimulation. Another study showed high specificity and a high positive predictive value for the prediction of poor response in IVF at this cutoff level (17).

Multivariate logistic regression analysis was used to study the relation among age, FSH, AFC, and ongoing pregnancy. Forward selection of parameters was applied with $P<.05$ for entry.

A total of 221 women were included in this study. In 9 patients, there was no basal serum FSH level available, and in 12 patients, basal FSH was determined in another hospital. In 36 patients, the basal FSH level was determined >1 year before the start of the IVF stimulation. In these cases, basal FSH was thought to be not representative anymore for the ovarian reserve status at the time of IVF treatment initiation. These 57 patients were excluded from the multivariate regression analysis and from the comparison of the outcome measures between patients with normal and elevated basal FSH levels, leaving 164 patients eligible for this evaluation. For the evaluation of AFC as a predictor of pregnancy, all 221 patients were analyzed.

The majority of the patients underwent a conventional IVF treatment (n = 204). Seventeen patients had an indication for an IVF–ICSI procedure. The main indications for IVF treatment included tubal factor (46 patients, 21%), male factor (88 patients, 40%), and unexplained infertility (87 patients, 39%).

When patients were analyzed by categories of normal (<15 IU/L) and elevated basal FSH levels (≥15 IU/L), there was a significant difference in normal response rate but not in clinical and ongoing pregnancy rates (Table 1). Patients also were classified on the basis of AFC (<5 and ≥5 follicles). Patients with a normal AFC not only had a significantly higher normal response rate but also had significantly better pregnancy rates. The AFC correlated neg-

TABLE 1

Ovarian response and pregnancy rates stratified by basal FSH level and antral follicle count.

Parameter	FSH <15 IU/L (n = 138)	FSH ≥15 IU/L (n = 26)	*P* value[a]	AFC ≥5 (n = 155)	AFC <5 (n = 66)	*P* value[a]
Normal response	68 (60–76)	27 (10–44)	<.01	78 (72–85)	33 (22–45)	<.01
Clinical pregnancy rate	22 (15–29)	15 (2–29)	.46	28 (21–35)	11 (3–18)	<.01
Ongoing pregnancy rate	13 (7–19)	12 (0–24)	.83	18 (12–24)	6 (0–12)	.02

Note: Values are percentages (95% CI). AFC = antral follicle count.
[a] χ^2 test.

Klinkert. AFC is a better marker than basal FSH. Fertil Steril 2005.

atively with chronological age; the Spearman correlation coefficient was −0.28, which is significant at the .01 level. When we performed a multivariate logistic regression analysis, the AFC was the only variable selected (odds ratio, 1.11; 95% confidence interval, 1.01–1.21; $P=.02$), meaning that the AFC is significantly associated with the occurrence of an ongoing pregnancy after IVF treatment. Age and basal FSH were not selected. After a forced entry of age into the prediction model, the AFC was still selected, meaning that the AFC significantly improved the prediction of ongoing pregnancy, given the age of the patient.

Seventy-one of the 113 patients aged >41 years had a normal AFC (63%). These patients, who are normally not treated in our clinic because of an assumed poor prognosis, had an ongoing pregnancy rate of 20% per cycle. However, the oldest patient to become pregnant in this study was 43 years old. None of the 31 women older than this age conceived, even though 13 of them had a normal AFC. Exclusion of these patients from the analyses did not change the results of this study; the AFC remaining the best predictor of ongoing pregnancy.

This study shows that a substantial number of the women who normally are not accepted for IVF treatment because of their age still have acceptable pregnancy rates after IVF. With the performance of an AFC before the start of the IVF stimulation, older women with still-acceptable pregnancy prospects and women with less favorable outcome for IVF treatment can be distinguished.

So far the prognostic value of the AFC with respect to the occurrence of pregnancy in IVF has been limited. Only Nahum et al. (18) showed that the AFC was a better predictor of (clinical) pregnancy than are age and basal FSH. The present study shows that the AFC is a valuable test that can be used in older women to assess the individual chance of pregnancy in IVF. This finding is in contrast to those of other studies evaluating the use of the AFC as a predictor of pregnancy in IVF (6, 19). This might be due to the fact that we only studied patients aged >38 years. Younger poor responders in IVF may still have reasonable pregnancy rates because only the quantity and not the quality of the oocytes is diminished (4). This might explain why in these patients the AFC is a good predictor of response, but not of pregnancy, whereas in older patients, a low AFC reflects a decline in both quantity and quality of the oocytes. In these patients, a low number of antral follicles therefore implies poor pregnancy prospects.

In the present study, patients aged >38 years with an AFC of less than five have poor pregnancy prospects. This is in line with the outcome of the studies performed by Chang et al. (19) and Kupesic et al. (8). Such patients should be properly counseled, probably against further treatment. In contrast, patients with a normal AFC have acceptable pregnancy rates. In fact, results for women aged younger than and older than 41 years did not differ. Therefore, we think that denial of treatment to patients aged ≥41 years is not justified anymore. The fact that patients aged ≥44 years failed to conceive in this study is consistent with the IVF results in older women that are presented in literature (2, 20, 21). Treatment of these women is not useful, not even when the AFC is sufficient.

The present study indicates that the AFC is a better marker than age and basal FSH for distinguishing between older patients with good and poor pregnancy prospects after IVF. Patients aged ≤44 years with a normal AFC still have acceptable pregnancy rates after IVF and therefore should not be denied treatment.

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