

REVIEW

How Can We Predict Success in Poor Responders?

Alina Elena BORDEA^{1,2}, Elvira BRATILA^{1,2}, Diana MIHAI^{1,2}, Marina ANTONOVICI³, Claudia MEHEDINTU^{2,3}, Andreea CARP-VELISCU^{1,2}

Abstract

Objectives: The aim of this study was to evaluate how can we predict success in poor responder patients in terms of pregnancy rate and live birth rate. **Material and method:** This study is a review of the newest papers that have in the center the poor responders undergoing treatment involving assisted reproductive techniques (ART). **Outcomes:** The results show that the most reliable factors when counseling a poor responder patient are age and Anti-Müllerian hormone (AMH) level. **Conclusions:** The most important factors that influence pregnancy rate are age and ovarian reserve, but other factors such as male pathology and laboratory techniques must be studied deeper.

Keywords: poor responders, AMH, age, pregnancy rate, live birth rate.

Rezumat

Obiective: Obiectivul studiului este evaluarea mijloacelor prin care putem prezice succesul la pacientele cu răspuns ovarian scăzut la stimularea ovariană controlată din ciclurile de FIV. **Material și metodă:** Această lucrare este un review al literaturii de specialitate recente. **Rezultate:** Rezultatele arată că cei mai buni predictorii ai ratei de sarcini sunt vârsta și nivelul seric al hormonului anti-Mullerian. **Concluzii:** Cei mai importanți factori care influențează rata de sarcini sunt vârsta și rezerva ovariană, dar alți factori precum patologia masculină sau tehnicile de laborator ar trebui mai amănunțit studiați.

Cuvinte cheie: pacientele cu răspuns ovarian scăzut, AMH, vârsta, rata de sarcini, natalitatea.

¹„Prof. Dr. Panait Sârbu” Clinical Hospital of Obstetrics and Gynecology, Bucharest, Romania

²„Carol Davila” University of Medicine and Pharmacy, Bucharest, Romania

³ Department of Obstetrics and Gynecology, „Nicolae Malaxa” Clinical Hospital, Bucharest, Romania

Corresponding author:

Claudia MEHEDINTU, „Carol Davila” University of Medicine and Pharmacy, Bucharest, Romania
E-mail: claudiamehedintu@yahoo.com

INTRODUCTION

One of the most frequently addressed questions to the infertility specialist, before a couple decides to use in vitro fertilization (IVF) to conceive, is: "What are the odds of getting pregnant through IVF?". In order to be able to answer this question correctly, many studies have tried to graphically show the real pregnancy rate. Most graphs consider the most important parameters in assessing the chances of obtaining through IVF a pregnancy finalized with an alive newborn, namely: age and ovarian reserve¹.

If for many causes of infertility there are variants of targeted treatments and therapies, for age and ovarian reserve that decreases with aging, there is still no effective treatment².

In recent years, the birth rate has increased among women over the age of 30 and up to 44 years. Delaying the conception of a child has multiple causes: professional and educational goals, ensuring economic status before birth and so on³.

The POSEIDON (Patient-Oriented Strategies Encompassing Individualized Oocyte Number) group classifies poor prognosis patients with confirmed or expected inappropriate ovarian response to exogenous gonadotropins. The classification takes into consideration 3 main criteria: age, anti-Mullerian hormone (AMH) and the response to a previous controlled ovarian stimulation⁴.

More recent studies show that the measure of success is given by the number of euploid blastocysts we can obtain. At the age of 28, 3 blastocysts are needed to have a 90% chance that one of them will be euploid⁵. A 2021 study shows that for a 95% pregnancy rate, 3 euploid blastocysts are needed, independent of the age of the mother⁶. All true, but the number of euploid blastocysts we can get from a full IVF cycle is age-dependent, as many studies show. The pregnancy rate is also age dependent⁷.

OVARIAN FOLLICULOGENESIS AND OVARIAN AGING

Follicular development begins in the fourth month of intrauterine life. Primordial germ cells migrate from the endoderm of the yolk sac to the gonadal ridge, process during which they undergo mitotic divisions. Once reaching the gonadal ridge, the oogonia enter the first stage of meiotic division and become primary

oocytes. At 20 weeks of gestation, the number of follicles that form the ovarian reserve is about 7 million. Follicular atresia begins during the intrauterine life and continues until menopause. The number of germ cells decreases to about 2 million at birth, and only 300.000 remain by the time the menarche occurs, meaning that 94% of the initial follicles disappear by then⁸.

Follicular maturation is classified into 3 stages:

1) The stage of primordial follicles - represents the beginning of follicular development and takes place inside the maternal uterus. At this stage the immature oocytes are surrounded by a single layer of flattened granulosa cells. The follicles do not have vascularization, but feed by diffusion through GAP-type junctions. The transport of steroid precursors is also done by diffusion.

2) The stage of primary follicles - is characterized by the transformation of flattened granulosa cells into cuboidal cells and the increase in the number of cell layers. The diffusion of nutrients and cellular signals received from cells with gonadotropins receptors is also done with the help of GAP junctions. During this stage, the secretion of the pellucid area at the level of the oocyte occurs.

3) The stage of antral follicles - is characterized by the formation of a single antral cavity filled with follicular fluid, between the granulosa cells that form the cumulus oophorus (surrounds the oocyte) and the mural granulosa cells (surrounds the antrum).

Follicular development is considered independent of gonadotropin stimulation until the primary follicle consists of 4 layers of granulosa cells⁹.

The size of the ovarian reserve from the beginning and the percentage of follicles that will become atretic are genetically determined factors. This is also the case for the growth initiation rate. Therefore, the age of climax onset varies from patient to patient and has genetic determinism. Another argument that supports this is that there is a family association regarding the age of menopause for mothers and daughters¹⁰.

With advancing age, fertility decreases due to follicular exfoliation and oocyte quality. The rate of follicular loss doubles when the number of oocytes falls below 25.000. This happens around the age of 37.5¹¹.

Menopause is the final step in the ovarian aging process. The follicular decrease with age determines the occurrence of irregular menstrual cycles (due to increased levels of FSH – Follicle-stimulating Hormone) and finally the cessation of menstruation. The parallel

decrease in oocyte quality contributes to the gradual decline of fertility and ultimately to natural sterility.

FSH is the initiator but also the finisher of folliculogenesis in the human ovary. Significant increases in circulating FSH levels are caused, through negative feedback, by decreased levels of follicular growth factors and follicular differentiation factors related to b-growth transformation factor. AMH is produced by immature ovarian follicles and helps regulate FSH secretion from the pituitary gland. As the number of immature follicles decreases with aging, the negative feedback on FSH decreases and thus the circulating levels of FSH increase. Increased circulating levels of FSH cause improper maturation of granulosa cells of preantral follicles, which have not completed their gonadotropin-independent growth cycle. These follicles will become atretic due to the asynchronous maturation of the somatic and germinal components¹².

The difference in ovarian aging among women is evident by the difference in age at which menopause occurs. Recognition of women who have low ovarian reserve for their age is extremely relevant for clinical practice. Ovarian reserve assessment tests are relatively accurate in evaluating the response to ovarian stimulation and in evaluating the chances of conceiving through IVF techniques¹³.

INFLUENCE OF AGE ON PREGNANCY RATE

Age is the main determinant for success in IVF. Fertility decreases naturally with age in both natural and ovarian-stimulated cycles. The decline in fertility begins around the age of 30 and the slope becomes steeper after 35 years. The live birth rate is inversely proportional to the mother's age⁷. Moreover, the rate of miscarriage and aborted pregnancies is directly proportional to maternal age, largely due to aneuploidies that increase in frequency with advanced maternal age¹⁴. Of course, low ovarian reserve is also a factor in predicting pregnancy rates, but even in these cases age is the main determining factor. In other words, women with low ovarian reserve and under the age of 35 have better pregnancy rates than those over this age¹⁵.

Maternal age is the leading cause of embryonic aneuploidy. In other words, the success rate to obtain a euploid blastocyst, able to give a pregnancy and also the implantation rate decrease significantly over the age of 40 years¹⁶.

The chances of a blastocyst being euploid decrease each year. If the rate of euploid blastocysts is about 60% in women under 35, for women aged 40-42 the rate of euploid blastocysts is a maximum of 30%. In other words, at the age of 28, 3 blastocysts are needed for at least one of them to be euploid (90% probability). For ages 35, 37, 39, 41, 43 and 45, 4, 5, 6, 9, 16 and 29 blastocysts are needed for one to be genetically correct⁵. As mentioned before, the rate of aneuploidy increases significantly after 30 years. The lowest rate of aneuploidy is found in the 26-30 age group¹⁷. More than 90% of these imbalances are of maternal origin caused by incorrect chromosomal segregation during oogenesis¹⁸.

However, predicting the success rate based on ovarian reserve alone is not enough. It is also necessary to correctly assess the ovarian reserve before approximating the chances of obtaining a pregnancy¹⁹.

INFLUENCE OF OVARIAN RESERVE ON PREGNANCY RATE

1. Definition of ovarian reserve

The concept of ovarian reserve describes a woman's natural endowment and is in close relation with age. The size of the follicular pool in a woman's ovaries between birth and menarche symbolically reflects the starting point of follicular decline. As we already know, most follicles are lost from intrauterine life. The follicle pool varies in size depending on genetically determined factors. If the average number of follicles at birth is around 300,000 per ovary, at the time of menarche this number is significantly lower²⁰.

The recruitment rate is also variable: between 100 and up to 7.500 follicles enter the process of maturation and growth each month. The peak is reached around the age of 14. The number of recruited follicles decreases with age regardless of the initial size of the ovarian reserve^{21,22}.

In other words, fertility decreases with age. This is not only caused by the decrease in the number of follicles, but also by the decline in oocyte quality. The rate of follicular loss becomes 2 times faster when the number of oocytes reaches below 25,000. This happens around the age of 37.5 years¹¹.

Therefore, the ovarian reserve is defined as „the number of oocytes left in the ovary”. The amount (number) of oocytes left in the ovary (follicular pool) differs from the oocyte quality. The latter reflects „the potential of a fertilized oocyte to give birth” 21.

As previously described, the decline in female fertility (decreased ovarian reserve) simultaneous with age is well known. Of course, there can be quite large differences between the ovarian reserve of several women of the same age, as this is genetically determined. In other words, the size of the ovarian reserve and the loss of the ovarian reserve as we age are genetically determined⁹.

2. Measuring ovarian reserve

Ovarian reserve assessment tests have become part of standard clinical practice because they can predict ovarian response during controlled ovarian stimulation. They allow patients to be informed about the expectations they should have²³⁻²⁵.

Measurement of ovarian reserve involves biochemical tests and imaging tests (transvaginal ultrasonography of the ovaries). These markers can estimate the number of oocytes we can extract following controlled ovarian stimulation and ovarian puncture for IVF. When assessed independently of age they are not as effective in predicting reproductive potential²⁶.

A large number of studies have investigated the role of the antral follicle count (AFC) in predicting the ovarian response to controlled ovarian stimulation. Thus, the 2019 ESHRE (European Society of Assisted Human Reproduction and Embryology) guideline concluded that predicting the ovarian response to controlled ovarian stimulation based solely on AFC is a reliable marker¹⁹.

Moreover, the AFC prediction in terms of pregnancy rate is a good one²⁷.

FSH is an intensely studied hormone when it comes to assessing ovarian reserve and response to ovarian stimulation treatment. Intracycle variability makes it most commonly measured during the follicular phase. FSH is frequently measured on days 1-4 of the menstrual cycle along with estradiol. An FSH above 10 mIU / ml is associated with a low response to controlled ovarian stimulation and low pregnancy rates²⁸.

In recent years, AMH has become the main marker for the evaluation of the ovarian reserve. It is produced by the granulosa cells of the preantral follicles and the small antral follicles. Thus, AMH reflects the exact size of the primordial follicular pool. Among the advantages of using AMH in the evaluation of the ovarian reserve are:

- it can be measured at any time during the menstrual cycle;
- it correlates well with the number of oocytes we can obtain from controlled ovarian stimulation;

- it correlates well with the rate of clinical pregnancies^{29,30}.

Tests to determine serum AMH levels have evolved from manual methods to more robust and automated methods. They have good analytical performance, including improved accuracy and a limit of quantification ≤ 0.08 ng / ml³¹.

To predict a poor response (≤ 4 oocytes collected after controlled ovarian stimulation), the cutoff value of the AMH level of 0.93 ng / ml was associated with sensitivity, specificity, positive predictive value and negative predictive value of 63.5%, 89.2%, 52.2%, respectively 92.9%²⁹.

The data from the literature are contradictory when we talk about the correlation of AMH with the live birth rate^{27,32}.

In patients with low ovarian reserve, AMH correlates with the age-specific rate of clinical pregnancies. When we take into account both age and ovarian reserve in patients with low ovarian reserve, the prediction of the clinical pregnancy rate is much better³³.

In patients with low ovarian reserve, AMH ≤ 1.05 ng / ml is associated with low live birth rates, in contrast, patients with AMH ≥ 1.06 ng / ml have much higher live birth rates. In patients under 35 who undergo an IVF procedure, serum AMH levels are an independent predictor of pregnancy rate³⁴.

Ovarian reserve assessment markers do not predict the likelihood of pregnancy during sperm insemination from the donor in unstimulated cycles. AMH does not predict pregnancy rate after ovarian stimulation and intrauterine insemination³⁵. Although positively correlated with each other and with IVF results, AMH and AFC can be discordant up to 30% of the time, as shown by Li H.W. et al. (2014) in his research³⁶.

The ESHRE Guide postulates the following¹⁹:

1. The prediction of the ovarian response by the value of AMH is reliable.
2. The prediction of the ovarian response by baseline FSH and no other ovarian reserve assessment marker is uncertain.
3. The prediction of the ovarian response by Inhibin B without another ovarian reserve assessment marker is uncertain.
4. The prediction of the ovarian response by estradiol value without another ovarian reserve assessment marker is uncertain.

The ESHRE guide recommends the use of AFC or AMH to the detriment of other tests when we need

to predict the response to controlled ovarian stimulation¹⁹.

AMH and AFC are important means of predicting the chances of success, but there are other variables that should not be neglected and that play a major role in the end result, namely: age, sperm quality (male pathology), genetic quality of embryos, stimulation protocols, laboratory procedures and techniques, embryo transfer technique^{6,37,38}.

CONCLUSIONS

Giving that fertility naturally decreases with age in both natural and ovarian-stimulated cycles, the decline starting around the age of 30 with a steeper slope after 35 years, and that nowadays women tend to delay procreation due to the prioritization of their career and financial status consolidation, ART are more and more used and infertility specialists try more often to bend the natural boundaries of fertility.

Although AMH has become the main marker for the evaluation of the ovarian reserve and is positively correlated with AFC and IVF results, it cannot predict pregnancy rate after ovarian stimulation and intrauterine insemination. Variables that should not be neglected and that play a major role in the end result are sperm quality, genetic quality of embryos, stimulation protocols, laboratory procedures and techniques, embryo transfer technique and further research is needed to evaluate how these variables influence pregnancy rate in poor responders.

Compliance with ethics requirements: The authors declare no conflict of interest regarding this article. The authors declare that all the procedures and experiments of this study respect the ethical standards in the Helsinki Declaration of 1975, as revised in 2008(5) and the national law. Informed consent was obtained from the patient described in the clinical case and his parents.

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