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## Immunological infertility

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### Abstract

We talk about infertility when a couple, despite regular intercourse 3-4 times a week, cannot get pregnant within a year. The problem of infertility equally affects both women and men. There are at least few reasons for this. A less common cause of infertility is immunological infertility. The paper presents the history of a couple affected by this type of infertility.

**Key words:** immune infertility, lymphocyte vaccinations.

## **Introduction**

We talk about infertility when a couple, despite regular intercourse 3-4 times a week, cannot get pregnant within a year.

Infertility in couples is an increasingly common problem, it is estimated that it affects even 10-18% of couples in the reproductive age, i.e. at the age of maximum professional, family and social activity. In Poland, the data shows about 10-15% of infertile partners, which translates into the number of about a million people unable to get pregnant. These are only estimates, as there are no detailed epidemiological studies - some couples will never try to obtain medical help [1-5].

Debates about infertility, whether it is a disease (or not), usually focus on the ethical and moral aspect of the treatment methods used. Recognizing infertility as a disease would have consequences in terms of the necessity to pay for funding by the state. Opponents of the concept of infertility as a disease raise arguments such as that the concept of "disease" should refer to a single entity, there should be periods of latency, harbingers and ending the disease, as well as signs and symptoms, local and general symptoms. Thus, infertility does not quite fit into this pattern, as its basic symptom is the lack of a child in a relationship. It does not cause physical pain and is not life threatening, it usually applies to a married couple, because often in other relationships each of the partners may have a child. The way to overcome (cure) infertility is to get pregnant and give birth to a healthy child [6].

The problem of infertility equally affects both women and men. There are at least four reasons for this:

- fallopian tube factor,
- endometriosis,
- ovulation disorders associated with polycystic ovary syndrome
- ovulation disorders of a different etiopathogenesis.

The main causes of male infertility are:

- oligoasthenospermia (34%),
- asthenospermia (29%),
- teratospermia (18%),
- azoospermia (9%),
- less frequently oligospermia or an immune factor.

Sometimes the cause of infertility is impossible to diagnose - then it is possible to presume poor quality of eggs, disorders of the sperm fertilizing ability and changes in the sperm's nuclear chromatin. The psychological factor is also considered, and research indicates that it may be the cause of 5% of infertility [7-11].

A less common cause of infertility is immunological infertility. According to estimates, immunological disorders may be the reason for difficulties in obtaining and maintaining pregnancy in about 10–15% of infertile couples. The paper presents the history of a couple affected by this type of infertility.

**Case study:**

The couple (now she is 35 and he is 40) started trying for a child in September 2015. The woman's hormones were tested, everything was normal, AMH was 2.74 ng / ml and was adequate for age. In March 2016, the monitoring of the cycle began, each time the egg follicle grew to an appropriate size, its rupture was supported by an injection of Ovitrelle. The results of the man's sperm were within the standards recommended by WHO, apart from the amount of properly built sperm - it was only 2%. However, the sperm count was 150 million (with WHO standards  $\geq 39$  million), so they were considered good. In May 2016, the woman had a fallopian tube patency test - hysterosalpingography. Both fallopian tubes were open.

In July 2016, the couple changed their gynecologist, he started cycles stimulated with clomiphene (Clostilbegyt), 4 mature egg follicles were achieved in the cycle at a time, their rupture was triggered by Ovitrelle. After three unsuccessful cycles, diagnostic laparoscopy and insemination were prescribed. Within 7 months, the couple underwent 4 unsuccessful intrauterine inseminations. In May 2017, the couple went to an infertility clinic, where idiopathic infertility was diagnosed and suggested in vitro. The man was not ready for this method of treatment, so the natural efforts continued until July 2018, where there were again two inseminations on the natural cycle. The woman's AMH dropped to 0.99 ng / ml. In December 2018, the couple returned to the infertility clinic, where preparations for the in vitro procedure began.

This was due to duplicate sperm results (morphology still 2%), virological tests. The woman was stimulated by menopurina, in March there was a pick up 36h after administering a double dose of Ovitrelle. 7 mature eggs were reached and time lapse was used to monitor the fertilization process. None of the cells have fertilized, not even the fertilization process has been initiated. In April 2019, in-depth semen tests were performed - semen test extended to MSOME classification, sperm nuclear chromatin integrity test and semen test extended to the HBA test. It turned out that the HBA result was 72% with the norm  $\Rightarrow$  80%. In May, the couple started the second in vitro procedure, at the couple's request, the IMSI (Intracytoplasmic Morphologically Selected Sperm Injection) procedure was applied, which consists in intracytoplasmic injection morphologically and qualitatively selected sperm.

With reduced HBA results, the pICSI method should be used, which is performed on special plates with hyaluronic acid. Binding with hyaluronic acid allows the selection of the correct sperm, which in many cases increases the percentage of correctly fertilized cells. Unfortunately, the couple's clinic did not have such a method of treatment. As a result of the stimulation, 6 mature cells were reached. In Poland, an attempt is made to fertilize 6 eggs as a standard, while in the third procedure and in a situation where no embryos were obtained in the previous procedure, it is possible to fertilize more cells. In this case, however, only 6 mature cells were obtained, of which two blastocysts of class 5.1.1 and 3.2.3 were obtained. During stimulation, it turned out that a polyp was formed, so the decision was made on the criotransphere.

In July 2019, hysteroscopy combined with scratching, i.e. deliberate and controlled damage to the endometrial membrane, was performed to increase the susceptibility of the endometrium to implantation. In August 2019, the 5.1.1 blastocyst criotransfer took place, Acard 75 mg and Neoparin 0 mg / 0.4 ml were used.

On day 10 after transfer, beta hcg level was 25 mIU / ml, on day 12 after transfer, beta hcg level dropped to 10 mIU / ml. In September 2019, a second class 3.2.3 embryo was administered, in addition to Acard and Neoparin, Encorton 5mg was used, the pregnancy lasted until the 6th week, there was a spontaneous abortion with bleeding. Doctors from the infertility clinic suggested a third in vitro procedure, but the woman started further self-diagnosis of the problem.

### **Self-diagnosis of the pair**

In October 2019, the results of the sugar and insulin curve were consulted again, this time with a gynecological endocrinologist. Hypoglycaemia and hyperinsulinemia were diagnosed, which could have a conceptual influence on the quality of the egg sacs. Previous doctors saw no abnormalities as the fasting results were normal. Metformin treatment (Siofor 500 mg x 2) was introduced, and a diet similar to insulin resistance was introduced. In addition, homocysteine, dhea (Dhea Eljot x 2 supplementation was implemented), vitamin D level, prolactin (hyperprolactinemia was found and Bromergone was introduced), ANA1, ASA, again TSH (patient with hypothyroidism) were tested. In addition, karyotypes were examined, the husband examined AZF and CTFR, the woman examined congenital thrombophilia (Fig. 1), PAI HOMO and both MTHFR hetero mutations came out, so heparin and Acard had to be implemented and heparin continued until the end of puerperium. Acard is usually discontinued by the end of the 33rd week of pregnancy. Supplementation with methylated folic acid was also introduced.

### **Immunological consultation**

The wife then searched for the term "autoimmune immunology" and, before going to the immunologist who deals with infertility, she examined KIRs (Kir BX came out, few implantation tubes missing), cytokines (Figure 2), and immunophenotype. In a cytokine study, the probable cause of infertility and two biochemical pregnancies after transfers emerged. The cytokines were completely shaky. Therefore, the couple also did a cross-match study, which showed that a partner can donate lymphocytes when treated with vaccinations.

In November, the couple went to Docent in Łódź, where they learned that with such results it would be difficult to keep the pregnancy because Interleukin 2 is very high (it destroys the embryo) and Interleukin 10 is definitely too low (it is responsible for the protection of the embryo). The optimal level for IL10 is a minimum of 1500 pg / ml, whereas in this case it was only 176 pg / ml. The ratio of IFN to IL10 is also important. The assistant professor ordered her husband's lymphocytes vaccinated and natural efforts. Vaccinations consist in taking 100 ml of blood from the partner (the donor can also be any other person, it is important that they are not related to the woman) and isolating lymphocytes from her, which are then administered in three places subcutaneously in each woman's forearm. These vaccinations modulate the woman's immune system and make the system less aggressive towards the embryo and fetus, thus preventing embryo-fetal rejection. Studies show that immunology works until the 12th week of pregnancy, when the placenta begins to form, and the risk of rejection by the woman's body decreases. The most common recommendations are three vaccinations, every three weeks each, followed by pregnancy after the second.

Analizowany gen	Zakres analizy/ lista mutacji	Uzyskany wynik		Nr sekwencji referencyjnej
Czynnik V (Leiden)	G1691A/R506Q	FV L_1691G	prawidłowy	NM_000130
Czynnik V (R2)	H1299R	FV R2_4070A	prawidłowy	NM_000130
Czynnik II (Protrombina)	G20210A	FIIP_20210G	prawidłowy	M17262
MTHFR*	C677T	MTHFR_677C-T Układ heterozygotyczny	nieprawidłowy	NM_005957.4
MTHFR*	A1298C	MTHFR_1298A-C Układ heterozygotyczny	nieprawidłowy	NM_005957.4
PAI-1**	4G/5G	PAI-1 4G Układ homozygotyczny	nieprawidłowy	NM_000602.4

\* reduktaza metylenotetrahydrofolianu

Figure 1 Congenital thrombophilia test

Ocena cytokin i współczynnika Th1 / Th2				
Interferon gamma	↓	186,0	pg/ml	2046,6 — 4738,0
TNF alfa	↓	1177,0	pg/ml	1208,3 — 4156,1
Interleukina 10	↓	176,0	pg/ml	695,6 — 3048,4
Interleukina 4		27,0	pg/ml	20,0 — 80,6
Interleukina 5	↓	14,0	pg/ml	20,0 — 59,8
Interleukina 2		28,0	pg/ml	20,0 — 62,5
Wskaźnik IFN/IL4	↓	6,9	ratio	36,7 — 175,0
Wskaźnik TNF/IL4		43,6	ratio	19,7 — 149,5
Wskaźnik IFN/IL10	↓	1,1	ratio	1,2 — 3,0
Wskaźnik TNF/IL10	↑	6,7	ratio	0,7 — 2,8

Figure 2 Cytokines

The first vaccination took place on December 30, 2019, the reaction was quite severe, but it only showed local symptoms such as pain, itching, burning and lumps. The second vaccination took place on January 20, 2020, monitored ovulation took place on January 27, and the woman had the first confirmed natural pregnancy in her life. The pregnancy turned out to be biochemical, the failure could have been caused by both immunological and other factors. The doctor recommended further vaccinations, considering that the first two may not be sufficient for this couple. The third vaccination has been postponed to 5/03/2020. The reactions were diminishing after each vaccination.

On April 27, 2020 monitored ovulation took place, on May 8, 2020 beta hcg was 38.5 mIU / ml and on the following days it grew normally, on May 21, 2020, the embryo's heartbeat was visualized, and on May 25, 2020, a pregnancy was performed with lymphocytes, from May 28, 2020 a gynecologist the attendant of pregnancy introduced (with the consent of the immunologist Docent) Encorton in a dose of 1 x 5 mg. As of today (August 19, 2020), the woman is in the 19th week of a properly developing pregnancy. Vaccinations were helpful for this couple. For many years it was impossible to achieve a natural pregnancy, which changed after the second vaccination.

### Summary:

Vaccination with partner's lymphocytes is considered a controversial method in Poland. They raise questions about possible side effects in the future, according to the instructor, the only side effects may be felt when the woman will have to be a kidney recipient - then the transplant doctor should be informed that such treatment took place in order to select the appropriate doses of immunosuppressive drugs. Each trying couple should consider for themselves to what extent the possible side effects outweigh the benefits of pregnancy.

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