

Assisted reproduction techniques (ART) – yesterday and today

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Abstract

The main reference to all medical activities is their undertaking in accordance with the principles of evidence based medicine-EBM.

The treatment of infertility with assisted reproduction techniques is subject to the same principles and, together with the dynamic progress of knowledge and new methods of treatment, requires ongoing regulation by scientific and medical bodies and the extension of competences of the clinics applying them.

The article below reviews the currently used ART reproduction techniques, with reference to the history of their creation, development and current new therapeutic solutions in this field.

Key words:

infertility, assisted reproduction techniques, insemination, in vitro fertilization

The concept of assisted reproduction techniques (ART) encompasses a medical action that bypasses one of the natural stages of fertilization, bringing the possibility of having a child to couples in whom the natural conception according to the diagnosis is impossible. Traditionally, these techniques include insemination with the sperm of a partner or donor, controlled multiple ovarian follicles stimulation with their collection and in vitro fertilization and embryo transfer [1,2,4].

The principles of good medical practice in relation to assisted reproduction technologies assume that these will be highly specialized diagnostic and therapeutic procedures used in specialized centers by qualified medical staff. In order to ensure not only high efficiency but also safety of treated patients, appropriate expert groups are issuing current recommendations and develop standards of conduct[1,4].

Apart from the strive for high quality and effectiveness of infertility treatment, it is recommended to provide patients with the maximum safety in the place of the activity, minimize the number of complications such as ovarian hyperstimulation syndrome (OHSS), continue to optimize the results of treatment e.g. by limiting the number of multiple pregnancies [1].

Periodically, the reports of clinics operating e.g. in Europe indicate a steady upward trend in the number of ART cycles to be performed, a plateau in the use of the ICSI procedure, the number of clinically-obtained pregnancies and a continuous downward trend in the use of more than two embryos for transfer [3].

Intrauterine insemination

Insemination procedures carried out in humans were preceded by experiments conducted on breeding animals. The first references come from the 14th century from Arab countries where the first insemination on horses were made. John Hunter is considered to be a pioneer of this technique in humans, who conducted the insemination that ended with conception in 1785. In 1945 and 1955, the British Medical Journal reported pregnancies of frozen semen [5].

The classic indications for insemination remain: male factor, endometriosis, infertility with

an unexplained cause, ejaculation disorders [6]. In terms of semen placement during the procedure, intrauterine insemination – ICI – intracevical insemination, intrauterine intrusion – IUI – intrauterine insemination and intrauterine insemination - FSP-fallopian sperm perfusion are distinguished. The intrauterine insemination is most often used in clinical practice. [5,6].

The expected benefit from performing insemination is the transfer of a certain number of normal sperm cells with optimal mobility near the egg cell. The insemination procedure, when in the hands of an experienced doctor, is easy to do. Besides, it seems that it is non-aggravating for patients. All this makes insemination still most often recommended to patients struggling with infertility[5,6,7].

The effectiveness of the procedure in the cycle is assessed differently, but does not usually exceed 20% per treatment cycle. The important factors that increase the chances of success are ovulation stimulation prior to insemination and a proper semen preparation.[7,8,9]

In the process of qualifying a couple for insemination, it is important to confirm the patency of the fallopian tubes in the woman and to exclude the current inflammation. In the case of a male it is crucial to examine semen to exclude azoospermia or severely weakened parameters of the semen.[7,8,9]

One of the newest data from the French centers indicates the effectiveness of insemination measured by the live birth rate at the level of 8.4 to 17.6%. One of the main factors with a statistically significant effect on the time to pregnancy was the number of the mature follicles recruited as a result of stimulation of ovulation before the procedure of insemination (9.4% in the case of one compared to 15.2% for two follicles)[7].

The significant factors that increase the chances of success are pre-insemination controlled ovulation stimulation and appropriate semen preparation.

The general standards for the evaluation and preparation of semen contained in the laboratory guidelines of the World Health Organization (WHO2010) are widely known, but recent data indicate that most centers still use their own methods and materials [9]. Publications on the effectiveness of IUI have revealed

significant differences in procedures prior to insemination e.g. in semen preparation techniques, the interval between donation and preparation and the administration of semen.

An attempt to develop an optimal IUI treatment protocol based on the clinical data collected in this way, in the case of non-compliance with standardization, becomes impossible and affects the published inter-laboratory differences in the frequency of pregnancies [10].

Recently published results indicate the lack of a negative effect on the pregnancy rate resulting from a possible daily delay in giving a woman partner's spermatozoa, counting from the donation of sperm. This can be an alternative procedure for couples whose presence on the day of ovulation is impossible, and also it allows for the planning of different laboratory work [11].

Due to the fact that the majority of pregnancies after insemination occur within 4 treatment cycles, this treatment is recommended for no more than 6 therapeutic cycles [8].

One of the possible complications after the insemination procedure is pelvic inflammatory disease (PID). According to the latest data, this complication occurs with a frequency not exceeding 16 in 1000 patients, which results in a clinical recommendation that there is no indication for routine antibiotic preventive treatment after this procedure [12].

In vitro fertilization

The history of in vitro fertilization dates back to the end of the 19th century, when in 1890 prof. Walter Haepe from the University of Cambridge conducted the first known rabbit embryo transplantation.

In 1959, M.C. Chang was the first to deliver mammals - a rabbit after the IVF procedure. He published this information in Nature [13].

In 1965, at the John Hopkins Institute in the USA, Robert Edwards and Howard Jones reported the first successful fertilization of the human oocyte [14], yet it was not until 1973 that the first confirmed pregnancy after the IVF procedure was achieved. However, it ended with an early miscarriage [15]. Robert

G. Edwards himself, on October 4, 2010, was awarded with the Nobel Prize in the field of medicine and physiology for the development of the method of in vitro fertilization.

On July 25, 1978, in Oldham, England, the birth of the first child conceived by assisted reproduction techniques took place [16]. In Australia, this was achieved two years later in 1980, and in the United States in 1981.

The first description of clomiphene stimulation appeared in the year of birth of the first IVF child. In 1981, some publication appeared showing the simultaneous use of clomiphene and hMG in the stimulation protocol for IVF [17]. Subsequently, in 1982, a paper was published, containing information on the use of GnRH agonists to avoid premature luteinization and to increase control over stimulation [18].

The first pregnancy obtained by the oocyte donation technique in a patient after bilateral ovariectomy in an artificial cycle occurred in 1983 [19]. In the same year, a paper was published about the first successful delivery of a baby from a pregnancy obtained after the transfer of a frozen embryo [20].

Another significant achievement from the medical point of view that happened in 1983 was the first pregnancy, which resulted in the birth of a healthy child with the use of the IVF procedure with sperm obtained in a patient with obstructed deferent duct after surgical sampling from the testicles.

The first use of sperm microinjection into egg cells in a mouse was successfully completed in 1987. Interestingly, one of the observations of the team of researchers was the fact of obtaining more female fetuses than the male ones [21]. However, it was only in 1992 that the first successful ICSI procedure was performed on human gametes [22].

It needs to be mentioned that as early as in 1990, there were reports of the possibility of performing the procedure of incision of the human embryo (Assisted Hatching) [23]. In the same year, there appeared a publication informing about the possibility of using the GnRH agonist instead of hCG as a trigger in the oocyte stimulation procedure [24].

In 1992, information about the successful stimulation and transfer of the embryo in a patient stimulated with recombinant FSH appeared - presented

simultaneously by two teams [25, 26]. In 1994, the medical world heard about an effective patient's stimulation using highly purified FSH [27].

Among the successive achievements of the reproductive medicine deserving mentioning, there was the birth of a healthy child after the use of ICSI technique with previously frozen oocytes carried out by a team of prof. Porcu [31]. In 2000, the team of the same professor led to the birth of the first child conceived using frozen semen as well as oocytes [28].

In 2002, the pre-implantation genetic diagnosis (PGD) was carried out with subsequent transfer of examined blastocysts, from which healthy twins were born [29].

At the end of this short summary of the history of IVF, in which undoubtedly numerous, sometimes equally important milestones in the development of IVF techniques needed to be omitted, the delivery of the first child after maturation of oocytes in laboratory conditions (IVM - In vitro maturation) fertilized with male reproductive cells obtained from TESE - Testicular Sperm Extraction should be mentioned [30].

The classic IVF technique is currently used less and less frequently. It is being replaced by newer techniques such as ICSI or pICSI.

The classic IVF method consists in placing egg cells collected during puncture in a special plate, to which after about 3-5 hours, prepared sperm in a concentration of approximately 100,000 sperm per 1 ml is added. The evaluation of fertilization is carried out about 18-20 hours later. The presence of the male and female pronucleus proves proper fertilization. From now on, the process of active division of embryo cells begins, most often it lasts up to the blastocyst stage, i.e. 5 days, when the transfer of the hatching embryo to the uterine cavity is carried out.

The technique used in couples with previous failures of classical methods, in couples with less than 5 oocytes collected during ovarian puncture or when partner sperm parameters are abnormal (too low sperm count, abnormal motility) is ICSI (intra-cytoplasmic sperm injection). It involves the injection of a single sperm directly into the egg's interior using a glass "needle", the so-called micropipette. Cohort studies published in 2018 by Li et al. showed no

differences in LBR (live birth rate) and CLBR (cumulative live birth rate) between classic IVF method and ICSI technique (intra-cytoplasmic sperm injection) in couples approaching IVF for indications other than the male factor [13].

The indications for extending the ICSI technique with the pICSI (Physiological ICSI) method are a reduced result of the sperm binding test with hyaluronic acid (HBA) (less than 65%), repeated IVF and ICSI procedure failures, and asthenozoospermia or teratozoospermia.

In the ICSI method, the sperm before being injected into the oocyte is selected on the basis of motility and morphology. The pICSI method additionally enables the sperm selection on the basis of its ability to bind to hyaluronic acid - a feature that proves its maturity. In the work carried out by Avalos-Duran et al. there were no differences in LBR, CP (Clinical pregnancy), implantation and miscarriage rate between ICSI and pICSI in pairs qualified for IVF due to the male factor [14]. Other studies conducted by Erberella et al. showed a statistically significant increase in the biochemical and clinical pregnancies in couples subjected to the pICSI vs. ICSI procedure, however the size of the study groups was low [15]. A meta-analysis of randomized trials comparing both methods is necessary to show a statistical advantage of one method over another.

MACS (magnetic activated cell sorting) is a method of selection of sperm that enable the elimination of sperm cells that are apoptotic. This method is recommended in patients with high DNA fragmentation rate (%DFI > 15%), in couples with repeated failure of assisted reproduction techniques (improper division of embryos, high level of fragmentation, degeneration of embryos) or medical history with recurrent miscarriages. The results of the studies are not unambiguous. In the study published by Sheikhi et al. a higher percentage of fertilization was observed in the study group (using the MACS technique) in comparison to the control group. However, no statistically significant differences were found in LBR [16]. In a study conducted in 2018 by Ziarati et al. a higher percentage of high-quality embryos, clinical pregnancies and implantation rates were demonstrated in the group using the MACS technique [17].

Stimpfel et al. demonstrated that the use of MACS technique may result in more high-quality blastocysts in couples with a male factor, but only in the case of female partners over 30 years of age [18].

The differences in perinatal results resulting from the transfer of fresh and frozen embryos should also be mentioned. In a meta-analysis conducted by Wong et al. that included four randomised clinical trials analysing a total of 1892 women who underwent the IVF procedure and compared a freeze-all strategy with a conventional IVF / ICSI strategy, there were no statistical differences in CLBR found [19]. On the other hand, the meta-analysis carried out by Roque et al. showed that the transfer of frozen embryos compared to fresh transfer significantly improves clinical practice [20]. Basirat et al. analyzed 1014 cycles (426 transfers of fresh embryos and 588 transfers of frozen embryos) without statistically significant differences in PR (pregnancy rate) [21]. The meta-analysis of randomized trials conducted by Zhang et al. showed a higher percentage of LBR and CPR (clinical pregnancy rate) in the frozen-thawed embryo transfer group (FET) than in the fresh embryo transfer (ET) [22].

In addition, we should mention the work of Chen published in the NEJM indicating a higher percentage of LBR after the transfer of frozen embryos in patients with polycystic ovary syndrome [23].

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