

AN INTEGRATED APPROACH TO THE SELECTION OF EMBRYOS USING AUXILIARY REPRODUCTIVE TECHNOLOGIES IN INCREASING THE EFFECTIVENESS OF FETAL RESULTS

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ABSTRACT

Cryopreservation of gametes and embryos is one of the most pressing problems of modern reproductive medicine. A comparative analysis of the effectiveness of the use of natural and frozen materials was carried out in the embryological laboratory. With the help of carriers of the closed (n = 598) and open (n = 665) types, 1263 molten embryos were transferred. Most of the indicators were good when using open carriers. The data obtained allows us to conclude that vitrification of morphologically normal gametes does not reduce the effectiveness of cryodastures and maintains their reproductive status. The use of open-type carriers to vitrify and store embryos demonstrates high levels of embryo retention after cryoconservation, with rates of miscarriage and low loss. It is assumed that this is due to the direct contact of the biological object (embryo) with liquid nitrogen and high freezing levels, which can be a decisive factor in the successful determination of vitrification in open carriers.

Key words: assisted reproductive technologies, cryoconservation, human embryo, vitrification.

INTRODUCTION

Assisted Reproductive Technology (Yort) (Assisted reproductive technology (ART)) is a pre–and firmly entrenched method of preventing infertility in medical practice. In many cases, the expected result – effective termination of pregnancy and the birth of a healthy child – is achieved. However, even the most modern methods of hattoki do not allow you to pass the average 50-60% kiss of getting

pregnant after conducting the Yort program. Long-term treatments, treatments that patients have independently performed against infertility before resorting to Yort further reduce the chances of getting pregnant and having a healthy baby. In such cases, a demand arises to increase the effectiveness of Yort methodologies. The development of assisted reproductive technologies (Yort in the next place) requires a high level of cellular technology in the embryological stage. Cryoconservation of gametes and embryos is a clinically important method for increasing the cumulative frequency of pregnancy [1, 2, 6]. After the transfer is carried out in 60% of cases when conducting standard procedures that stimulate oocyte ovulation and fertilization in vitro conditions, freeze-fit embryos remain. As a result, cryoconservation provides an opportunity to continue treatment even if there is no pregnancy in the stimulation cycle. Frozen embryos can be thawed and transferred to the patient uterus in subsequent cycles, which does not require the use of expensive hormonal stimulation schemes for superovulation [2, 7, 8]. Another obvious advantage of cryoconservation is that if, as a result of the development of severe hyperstimulation syndrome in patient, there is also a risk of impaired implantation (bleeding, lack of secretory transfusion and endometrial polyps, as well as extreme difficulty in embryo transfer), it provides an opportunity to undeniably cancel embryo transfer in the stimulation cycle [3, 9, 10].

The purpose of the study. Comparison of the effectiveness of the use of natural and frozen gametes in cycles of assisted reproductive technologies and assessment of the impact of the carrier species on the results of cryodastures and cryoprotokoles for freezing and storage of embryos.

Research materials and methods. The study used oocytes, sperm and human embryos, the study of which was carried out following the international ethical and legal standards for working with human embryos [11-20]. The study of gametes and embryos was conducted from the base of embryological laboratories of the Zao "IDK medical company" as part of extracorporeal fertilization cycles. The use of sex cells and human embryos in scientific research is allowed by the Ethics Committee of the samara State Medical University of the Ministry of health of Russia. Gametes and embryos were identified under stereomicroscope control (Nikon, Japan). Sook (Australia) incubators were used to incubate under 5% O₂ conditions. Vitrolife (Swedish) environments were used to cultivate embryos up to 5-6 days of embryonic development. For embryo vitrification, Irvine Scientific (USA) environments and CryoTop (Japan) Open and Cryotype (USA) closed carriers were used. Cultivated 5-6-day blastocysts were evaluated according to the International Classification [4, 21].

In a computer statistical computing environment of results, R (R v.3.5.3, RStudio v.1.1.463) was statistically processed, with initial data input using MS Excel spreadsheets. Statistical methods, comparison tests of proportions, including accurate binominal for small samples and one-sample proportional tests correcting continuity for large samples, were used.

Research results and discussion.

An important element of the freezing process is the carrier in which the sample is located during the cryocostoration process and subsequent storage time [5,15]. For cryoconservation of oocytes and embryos, carriers must ensure that embryos are reliably stored during cooling, storage, and heating. Carriers are divided into two groups - open and closed. If the carrier is of an open type-the droplet of the embryonic cryoprotector solution is in direct contact with liquid nitrogen (e.g. Cryotope, Japan). In a closed carrier (e.g. Cryotope, USA), the cryoprotector solution does not come into contact with liquid nitrogen, and cooling is carried out through the protective sheath wall. The open and closed carrier has a protective sheath that prevents mechanical damage to the frozen embryo. To assess the effectiveness of different carriers, the following indicators are calculated: survival rate, average transplant coefficient of the embryo (s), average number of embryos transferred, pregnancy rate, birth and loss rate In 2016-2018, 1,263 embryos were dissolved in the carrier species under consideration. The survival of embryos in closed carriers was 82.6%, in the open state - 93.0%, and this difference is statistically significant ($r < 0.0001$). The average grafting score of Group I embryos was 3.9, and Group II - 4.1. The average number of transfer embryos is 1.2 and 1.3, respectively. Pregnancy rates varied significantly statistically ($r < 0.0001$) and were 36.3% (Group I) and 42.2% (Group II). The fertility rate of clinical pregnancy, given known results, was 72.5% in Group I, 62.1% in Group II, and losses were 25.1% and 22.3% respectively. Birth and loss rates varied significantly statistically. In general, the results of the study can be concluded that the survival rate of embryos is high when using open carriers during cryopreservation.

Conclusion. All the listed factors make it necessary to prioritize the development of modern technologies of embryological ETAP in the Yort program. The correct selection and application of embryonic acquisition, diagnosis and cultivation technologies leads to great progress in the work of auxiliary reproduction centers (1).

Vitrification of embryos makes it possible to maintain the quality and normal morphological and physiological state of the cells and make their use possible in the future. This makes it possible to apply this technology in addition to natural

materials, without reducing the quality and efficiency of the auxiliary programs of reproductive technologies. At the same time, open carriers applied to the freezing and storage of embryos demonstrate high indicators of the effectiveness of cryoprommas. Clinical indicators directly related to survival rates (frequency of clinical pregnancy, childbirth and fetal loss) also demonstrate the advantage of using open carriers from statistically different indicators in embryological practice.

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