ASSESSMENT OF THE EFFECT OF PGT-A ON THE FREQUENCY OF PREGNANCY OCCURRENCE DURING CRYOTRANSFER OF EMBRYOS IN A HYALURONAN-CONTAINING MEDIUM IN DIFFERENT AGE GROUPS

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Abstract

Relevance. Preimplantation genetic testing for aneuploidy (PGT-A) is widely used in assisted reproductive technology programs, but its clinical usefulness, especially in terms of increasing pregnancy rates, remains a subject of active scientific debate. With the growing use of embryo cryotransfer and adhesive media containing hyaluronan, assessing the contribution of PGT-A, taking into account the age factor, is becoming particularly important.

Objective. To evaluate the effect of PGT-A on the frequency of clinical pregnancy in frozen embryo transfer (FET) performed in a hyaluronan-containing medium in different age groups of patients.

Materials and methods. A comparative study was conducted on the outcomes of frozen embryo transfer (FET) in patients treated at the Israeli Medical Center (Tashkent, Uzbekistan) with and without preimplantation genetic testing for aneuploidy (PGT-A). All transfers were performed according to FET protocols using a hyaluronan-containing medium. Patients were stratified by age: under 35 years and ≥35 years. Two groups were formed: the PGT-A group (transfer of euploid blastocysts; n = 82) and the non-PGT-A group (n = 92). The primary endpoint of the study was the frequency of positive β-human chorionic gonadotropin (β-hCG+).

Results. The analysis revealed differences in the frequency of pregnancy between age groups of patients. Overall, the frequency of positive β-human chorionic gonadotropin (β-hCG+) in the PGT-A group was 51.21% (42/82), while in the non-PGT-A group it was 47.8% (44/92); the differences were not statistically significant ($p > 0.05$).

In the subgroup of patients aged ≥35 years, the frequency of β-hCG+ was 46.15% (22/52) in the PGT-A group and 40.0% (12/30) in the group without PGT-A. The data obtained indicate age-dependent variability in FET outcomes with no statistically significant differences between the groups.

Conclusion. The results obtained indicate the need for an individualized approach to the use of preimplantation genetic testing for aneuploidy (PGT-A) in cryotransfer of embryos, taking into account the age characteristics of patients. In FET conditions using a hyaluronan-containing medium, the transfer of euploid embryos after PGT-A was accompanied by a moderate tendency toward a higher frequency of biochemical pregnancy, especially in the age group ≥35 years, but the differences identified did not reach statistical significance. The data obtained emphasize the need for further studies with a larger sample size to clarify the clinical efficacy and determine the indications for the selective use of PGT-A.

Introduction

In recent years, frozen embryo transfer (FET) has become an integral part of assisted reproductive technology (ART) programs thanks to significant advances in embryo cryopreservation methods and improvements in endometrial preparation protocols. These achievements have contributed to higher implantation and clinical pregnancy rates, making the "freeze-all" strategy widely used in clinical practice. At the same time, preimplantation genetic testing for aneuploidy (PGT-A) is being actively introduced, aimed at selecting euploid embryos in order to potentially improve reproductive outcomes and reduce the risk of chromosomally-related reproductive losses.

Despite the widespread use of PGT-A, the question of its routine application remains the subject of active scientific debate. A number of studies and meta-analyses indicate a reduction in the incidence of spontaneous miscarriages and a reduction in the time to pregnancy when using PGT-A, while other studies do not show a statistically significant increase in the live birth rate, especially in patients of young reproductive age with a good prognosis. This highlights the need for a more differentiated approach to selecting patients for genetic testing of embryos.

An additional factor that may affect the success of implantation is the use of hyaluronan-containing adhesive media during embryo transfer. It is believed that hyaluronic acid improves the interaction between the embryo and the endometrium by enhancing cell adhesion, modulating the local immune response, and creating a more physiological microenvironment in the uterine cavity, which may be particularly important in FET programs.

Given the continuing ambiguity of clinical data, it seems relevant to conduct a comprehensive analysis of the impact of PGT-A on the outcomes of frozen embryo transfer, taking into account the age factor, as well as the use of hyaluronan-containing media. This approach will allow for a more objective assessment of the feasibility of combining these technologies and determine their role in improving the effectiveness of ART programs and live birth rates.

Materials and Methods

Study design. A comparative single-center study was conducted to evaluate the outcomes of frozen embryo transfer (FET) programs.

Patients. The study included patients undergoing infertility treatment at the Israeli Medical Center (Tashkent, Uzbekistan). Inclusion criteria were undergoing FET and the availability of data on the use or absence of preimplantation genetic testing for aneuploidy (PGT-A).

Stratification and group formation. Patients were stratified by age into two main groups: <35 years and ≥35 years. Each age group was further subdivided based on the use of PGT-A:

- PGT-A — transfer of euploid blastocysts following PGT-A;
- non-PGT-A — transfer of blastocysts without genetic testing.

Thus, the study included four subgroups:

1. <35 years, PGT-A;
2. <35 years, non-PGT-A;
3. ≥35 years, PGT-A;
4. ≥35 years, non-PGT-A.

The total number of patients in each main group was:

- PGT-A — n=82;
- non-PGT-A — n=92.

Embryological and clinical procedures. On Day 4 of development, embryos underwent mechanical assisted hatching using a microneedle to create a controlled opening in the zona pellucida. This approach facilitated trophectoderm herniation by Day 5–6 and improved access during biopsy, while avoiding potential thermal stress associated with laser-assisted techniques (Figure 1).

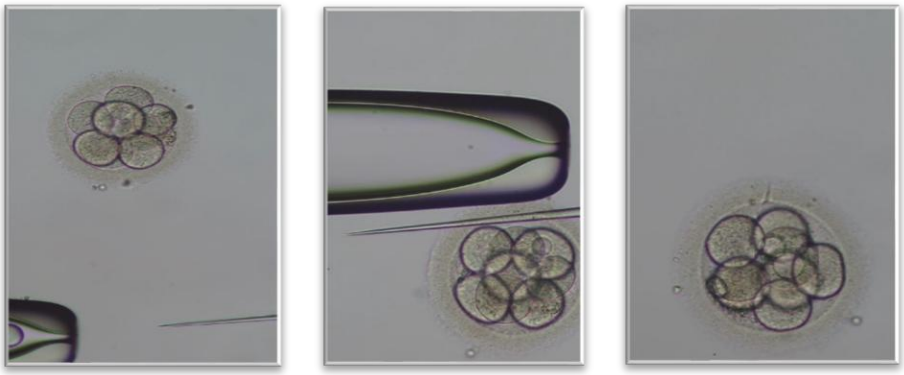


Figure 1. Mechanical assisted hatching of human embryos.

Blastocysts that had reached sufficient expansion on day 5 or 6 underwent biopsy using the "flicking" technique, developed to minimize mechanical trauma and preserve embryo viability. The procedure involved stabilizing the blastocyst with a holding pipette, aspirating the herniated trophectoderm cells through a mechanically opened pellucid zone, detaching 5–10 cells with a quick controlled flick motion, and immediately returning the embryo to culture for re-expansion (Figure 2). This method allowed a stable number of cells to be obtained while maintaining the structural integrity of the blastocyst. All embryos underwent NGS-based PGT-A prior to vitrification and were cryopreserved at the blastocyst stage.

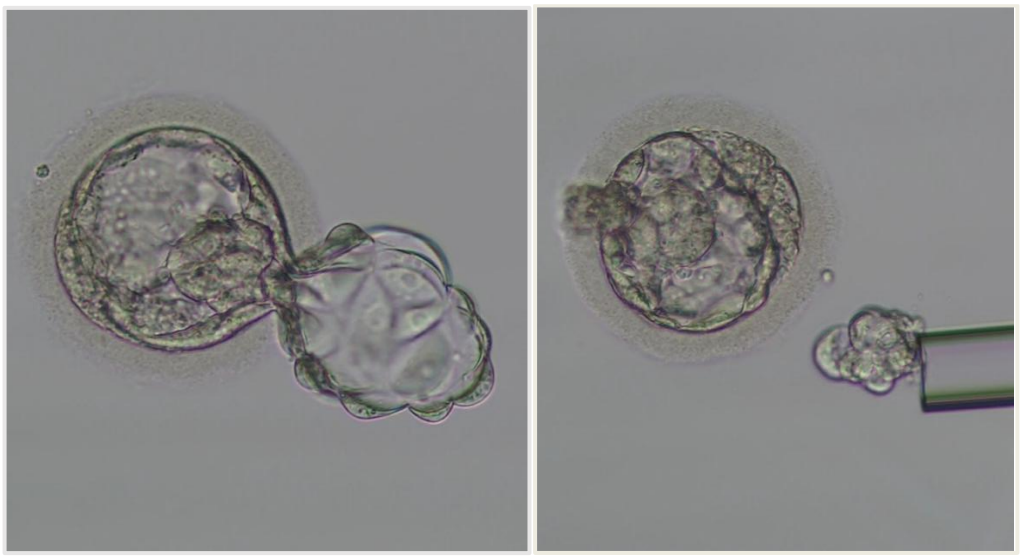


Figure 2. Trophoectoderm biopsy using the flicking technique.

Cryotransfer was performed according to FET protocols using a hyaluronan-containing medium. Endometrial preparation was performed according to standard clinical protocols of the clinic.

Outcome assessment

Assessment of results

The primary endpoint of the study was the frequency of biochemical pregnancies confirmed by the detection of positive levels of β-human chorionic gonadotropin (β-hCG) 14 days after frozen embryo transfer (FET). Clinical pregnancy was further confirmed by ultrasound examination performed two weeks after a positive β-hCG result, with visualization of the gestational sac and embryo in the uterine cavity.

Statistical analysis

Comparative statistical methods were used to analyze categorical data. Differences were considered statistically significant at $p<0.05$.

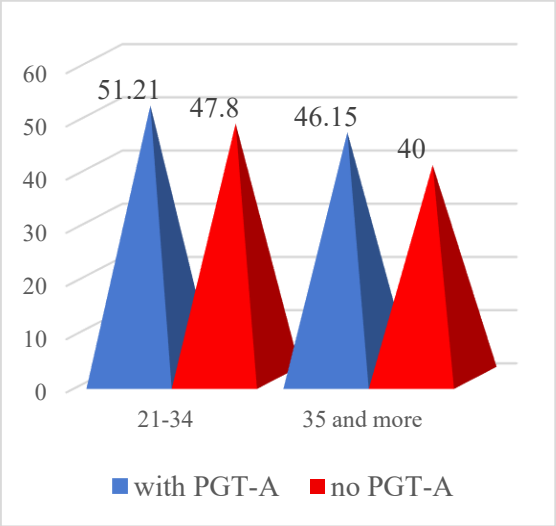
Results

PGT-A results. According to preimplantation genetic testing for aneuploidy (PGT-A), 68.4% of biopsied blastocysts were classified as euploid and selected for vitrification and subsequent transfer. The remaining 31.6% of embryos had chromosomal abnormalities, including aneuploid and mosaic forms, and were excluded from the analysis of clinical outcomes. Embryos with uncertain results were also excluded from further evaluation. Thus, the analysis of transfer outcomes was performed exclusively for genetically normal (euploid) embryos, which allowed us to exclude the influence of chromosomal status as a confounding factor.

Clinical outcomes taking into account age stratification. When analyzing the results taking into account the age of the patients, the following data were obtained.

In the 21–34 age group, in the PGT-A subgroup, frozen embryo transfer was performed in 82 patients. A positive β-hCG result was recorded in 42 women, which was 51.21%. In a similar age subgroup without PGT-A, 92 patients underwent embryo transfer, with a positive β-hCG detected in 44 women (47.8%). Intergroup differences in this age category did not reach statistical significance ($p>0.05$).

In the age group ≥35 years, the transfer of euploid embryos after PGT-A was performed in 52 patients, and a positive β-hCG was recorded in 22 women, which amounted to 46.15%. In the subgroup of patients of the same age without PGT-A, transfer was performed in 30 women, with a positive β-hCG result in 12 patients (40.0%). In this age group, there was also a tendency toward a higher frequency of biochemical pregnancy when using PGT-A, but the differences were not statistically significant ($p>0.05$).



Age	21-34 years		35+ years	
01.2024-12.2024	PGT	No PGT	PGT	No PGT
FET	82	92	52	30
Positive	42	44	22	12
Negative	40	48	28	18
Implanta-tion rate	51,21 %	47,8 %	46,15 %	40 %

Figure 3 and Table 1. Implantation rate (β-hCG+) after frozen embryo transfer (FET) with and without PGT-A in different age groups in % and abc indicators.

Overall assessment of results. Thus, in both age categories, the use of PGT-A was accompanied by a moderate tendency toward an increase in the frequency of biochemical pregnancy compared to transfers without genetic testing, which was more pronounced in patients aged ≥35 years. However, the absence of statistically significant differences indicates the need for cautious interpretation of the data obtained and emphasizes the advisability of an individualized approach to the use of PGT-A.

Discussion

The results obtained reflect the contradictory nature of the impact of preimplantation genetic testing for aneuploidy (PGT-A) on the outcomes of frozen embryo transfer (FET), which is generally consistent with the data in the current scientific literature. A number of large randomized studies and meta-analyses have shown that the use of PGT-A contributes to a reduction in the frequency of early reproductive losses and a decrease in the time to pregnancy, but its effect on live birth rates remains ambiguous, especially in the general population of ART patients. The absence of statistically significant differences in the overall sample of our study may be due to both limited statistical power and the influence of a number of confounding factors, including the morphological and functional quality of embryos, characteristics of endometrial receptivity, and endometrial preparation protocols in FET cycles.

Particular attention should be paid to the identified trend toward a higher frequency of biochemical pregnancies in patients aged 35 years and older after the transfer of euploid embryos. This fact is consistent with the idea of an age-dependent increase in the frequency of chromosomal aneuploidy in oocytes and embryos, which determines the potential clinical value of selective use of PGT-A specifically in older age groups. Previously published studies indicate that in women of late reproductive age, the use of PGT-A can improve implantation rates and reduce the risk of spontaneous miscarriage, whereas in young reproductive-age patients with a favorable prognosis, the benefits of this technology remain minimal or

statistically insignificant. This confirms the need for an individualized approach to the prescription of PGT-A, taking into account the age and reproductive prognosis of the patient.

At the same time, the data obtained do not confirm the advisability of routine use of PGT-A in all patients undergoing ART programs. Current professional recommendations emphasize that the unjustified use of PGT-A can lead to increased treatment costs, a potential reduction in the number of embryos available for transfer, and the risk of false-positive or mosaic results, the clinical significance of which remains a subject of debate.

The effect of hyaluronan-containing adhesive media used in embryo transfer should also be considered. Hyaluronic acid is known to be an important component of the extracellular matrix of the endometrium and plays a key role in cell adhesion and implantation processes. According to clinical studies, the use of hyaluronan-containing media may improve the interaction between the embryo and the endometrium, increasing the likelihood of implantation in both fresh and frozen transfer cycles. In our study, the use of this medium probably contributed to the optimization of implantation conditions in both groups, which could have leveled out the differences associated with the use of PGT-A.

Thus, the results confirm the need for a balanced and differentiated approach to the use of PGT-A in FET programs. The combination of genetic selection of embryos and optimization of transfer conditions, including the use of hyaluronan-containing media, appears promising, but requires further prospective studies with large samples to identify the groups of patients who can derive the greatest clinical benefit from these technologies.

Conclusion

The results of this study demonstrate age-dependent variability in the outcomes of frozen embryo transfer (FET) associated with the use of preimplantation genetic testing for aneuploidy (PGT-A). When using a hyaluronan-containing medium, the transfer of euploid embryos after PGT-A was accompanied by a moderate tendency toward a higher frequency of biochemical pregnancy in patients aged ≥35 years, but this difference did not reach statistical significance. The data obtained indicate a possible age-specific benefit of PGT-A in women of advanced reproductive age, while not confirming the advisability of its routine use in all patient groups. The results emphasize the need for an individualized approach to the use of PGT-A and indicate the advisability of conducting further well-designed prospective studies with an increased sample size and assessment of clinically significant outcomes, including the frequency of clinical pregnancy and live birth.

List of references

1. Gleicher N, Kushnir VA, Barad DH. *Preimplantation genetic testing for aneuploidy: a critical review.* Reprod Biomed Online. 2021;43(5):789–798. doi:10.1016/j.rbmo.2021.06.012

2. Munné S, Alikani M, Tomkin G, Grifo J. *Clinical outcomes after preimplantation genetic testing for aneuploidy in women of advanced maternal age.* Fertil Steril. 2020;114(2):321–330. doi:10.1016/j.fertnstert.2020.03.015

3. Forman EJ, Hong KH, Ferry KM, Tao X, Taylor D, Levy B. *Effect of PGT-A on implantation and pregnancy outcomes in frozen embryo transfer cycles.* Fertil Steril.

2019;111(6):1147–1155. doi:10.1016/j.fertnstert.2019.01.029

4. Gleicher N, Barad DH. *Are preimplantation genetic tests beneficial in young women?* Hum Reprod. 2022;37(7):1421–1430. doi:10.1093/humrep/deac067

5. Chen M, Qiao J, Zhao X, et al. *Hyaluronan-enriched transfer medium improves implantation rates in frozen-thawed embryo transfer cycles: a randomized study.* J Assist Reprod Genet. 2018;35:1231–1239. doi:10.1007/s10815-018-1265-7

6. Sunkara SK, Rittenberg V, Raine-Fenning N, Bhattacharya S. *Systematic review and meta-analysis of preimplantation genetic testing for aneuploidy (PGT-A) in IVF.* Hum Reprod Update. 2019;25(2):181–199. doi:10.1093/humupd/dmy038

7. Harper J, Harton G. *The ESHRE PGT Consortium: 10 years of PGT-A reporting and outcomes.* Hum Reprod Open. 2020;2020(2):hoaa018. doi:10.1093/hropen/hoaa018

8. Ata B, Kaplan B, Danzer H, et al. *Age-related outcomes in frozen embryo transfers with PGT-A versus conventional transfer.* Fertil Steril. 2021;116(1):88–97. doi:10.1016/j.fertnstert.2021.02.022

9. Yu Y, Li L, Liu P, et al. *Efficacy of hyaluronan-containing transfer medium on implantation rates in IVF: a multicenter study in Asia.* Reprod Biol Endocrinol. 2017;15:72. doi:10.1186/s12958-017-0291-9

10. Coates AJ, Forman EJ. *Clinical considerations in the use of PGT-A in IVF: benefits, limitations, and controversies.* Fertil Steril. 2022;118(4):651–661. doi:10.1016/j.fertnstert.2022.06.001