

# VALIDATION OF A RAPID VITRIFICATION AND WARMING PROTOCOL “FAST AND FURIOUS” FOR DONOR OOCYTES IN A CLINICAL IVF LABORATORY

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## Background

- Donor egg banks implement stringent morphological assessment criteria when selecting oocytes for cryopreservation.
- Oocytes exhibiting structural abnormalities—such as irregular polar bodies, prominent vacuoles, central cytoplasmic granularity, or abnormalities of the zona pellucida or oolemma—are excluded from clinical use.
- Although not suitable for patient treatment, these non-bank-eligible oocytes represent a valuable resource for laboratory validation and optimization of novel cryopreservation protocols.

## Objective

To assess the feasibility and performance of a rapid vitrification–warming protocol using previously vitrified–warmed donor oocytes with morphological abnormalities that were excluded from clinical egg banking.

## Materials and Methods

450 morphologically abnormal donor oocytes (*previously excluded from banking*) that are:

- Used for internal QC only
- Two-year period

Standard vitrification/warming



392 survived (87.1%)



Re-vitrified (*Fast & Furious – 2 minutes*)

Performed on a 90 mm dish lid at room temperature

- Rinse in Washing Solution (m HTF)
- 1 min – Equilibration Solution (7.5% EG/DMSO)
- 1 min – Vitrification Solution (15% EG/DMSO + 0.5 M sucrose)
  - Immediate Cryotop loading and LN<sub>2</sub> storage

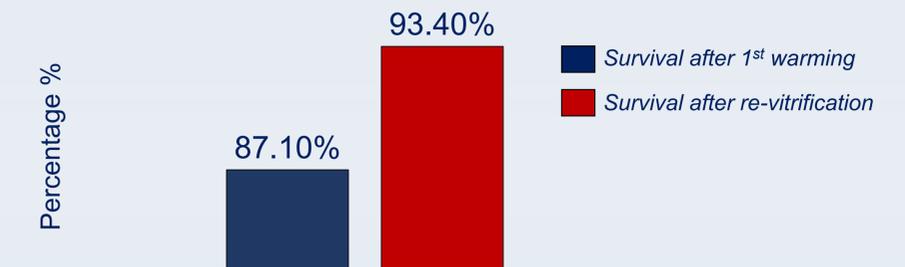


Rapid warming (*Fast & Furious – 2 minutes*)

- 1 min- Dilution solution (0.5 M sucrose) at 37°C
  - 1 min-Washing solution (m HTF) at 37°C
  - Recovery in global medium+ 20% protein

## Results

- Despite prior freeze–thaw exposure, all oocytes tolerated two vitrification cycles. Compared with standard warming protocols, there was no significant increase in oocyte rupture or observable structural damage. Post re-vitrification survival showed a percentage of 93.4%; a total of 366 oocytes survived.



## Conclusion

The protocol demonstrated high post-warming survival with reduced processing time and minimal handling. It shows promise for workflow optimization in clinical IVF laboratories. Rapid 2-minute vitrification protocol achieves 93.4% survival even after prior freeze-thaw exposure. However, further studies are required to evaluate its impact on fertilization rates, embryo development, and pregnancy outcomes.

## Limitations

- The study was limited to morphologically abnormal oocytes, which may not fully represent the response of clinically eligible oocytes and translational applicability remains uncertain.
- Fertilization outcomes were not assessed, precluding evaluation of downstream developmental competence.

## References

1. Wozniak, K., et al., *Ultra-fast vitrification and rapid elution of human oocytes: Part II - verification of blastocyst development from mature oocytes*. Reprod Biomed Online 2024. **49**(6): p.104690.
2. Liebermann, J., et al., *Fast and furious: successful survival and resumption of meiosis in immature human oocytes vitrified and warmed using a short protocol*. Reprod Biomed Online. 2024. **49**(1): p.103976.

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