POST-HATCHING VERSUS PRE-HATCHING BLASTOCYST BIOPSY FOR PREIMPLANTATION GENETIC TESTING (PGT) IS UNEXPECTEDLY ASSOCIATED WITH REDUCED IMPLANTATION

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Background: Single blastocyst transfer only recently became the norm for in vitro fertilization (IVF) in the United States. Before 2016, most IVF transfers were of multiple embryos. Multiple embryos were transferred in fewer than 20% of all US IVF cycles for the first time in 2020. Unlike multiple embryo transfer, single embryo transfer allows for complete and unambiguous data regarding outcomes of every transferred embryo. The newly standard practice of single embryo transfer thus provides unprecedented ability to reevaluate factors associated with successful embryo implantation.
Objective: To evaluate implantation according to oocyte source, blastocyst morphology, and cryosurvival among single vitrified euploid blastocyst transfers
Materials and Methods: All transfers (2021-2023) of single vitrified blastocysts previously diagnosed as chromosomally normal by next-generation sequencing (NGS) preimplantation genetic testing (PGT) at a single private fertility center were reviewed. Analysis was limited to embryos biopsied on Day 6 after oocyte retrieval, which constituted the bulk of transfers (76%). Blastocysts were graded by experienced embryologists according to Gardner's three-component system. Post-vitrification survival was graded as excellent, good, fair, or poor. Clinical pregnancy (gestational sac with ultrasound confirmed heartbeat) was evaluated by logistic regression analysis. Error bars indicate 95% binomial confidence intervals.
Results: Neither autologous patient age (p=.84) nor oocyte source (p=.14; donor n=1371 versus patient n=1128) significantly influenced pregnancy outcomes; these groups were therefore pooled in subsequent analyses.
Complete blastocyst grades were available for 2491 cycles. Most (79.5%) were biopsied at stage 3 or 4 (expanding/expanded), while 20.5% were biopsied at stage 5 or 6 (hatching/hatched). Inner cell mass and trophectoderm were more often graded A (58% and 57%) versus B (42% and 43%). Multiple logistic regression analysis indicated a clear association between these blastocyst grades and clinical pregnancy (model p<.001, McFadden's R^2=.01, area under the ROC curve =.546). Developmental stage (expanding/expanded versus hatching/hatched) was most predictive of pregnancy (β=.33, p=.001) followed by trophectoderm grade (β=.18, p=.033); inner cell mass grade was not significantly associated with pregnancy after accounting for developmental stage and trophectoderm grade in this multivariable analysis (β=.12, p=.16) (Figure 1). Figure 2 illustrates the additive combined effects of developmental stage and trophectoderm quality on pregnancy outcomes.

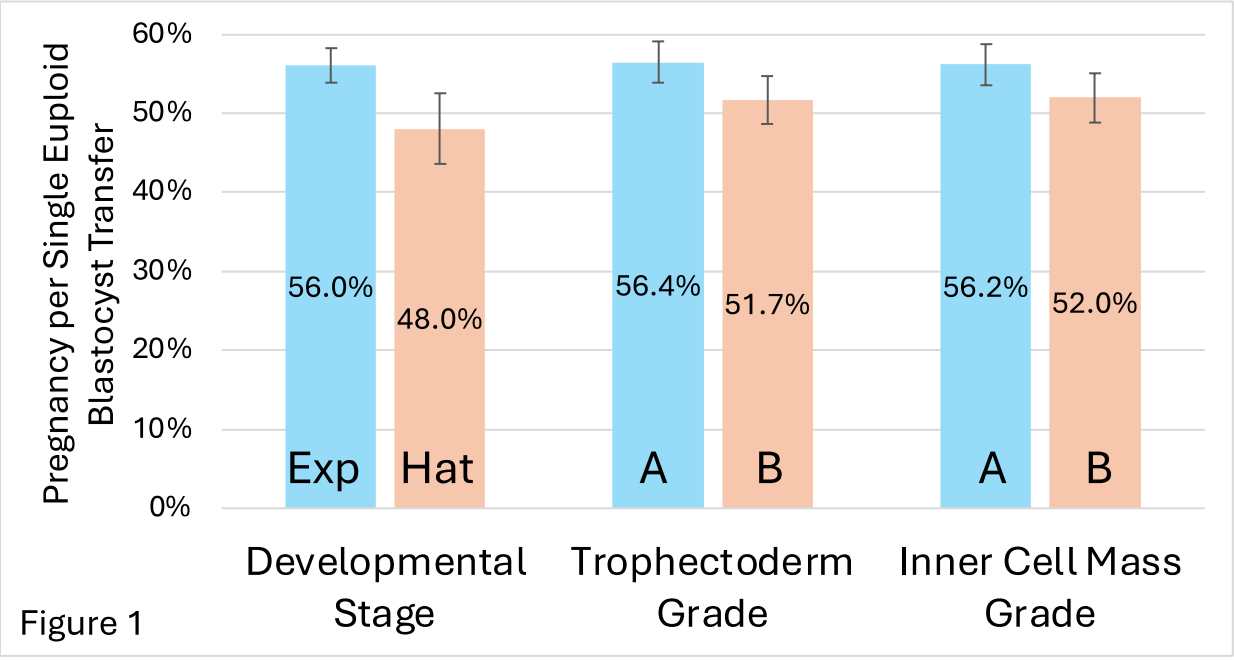

Figure 1

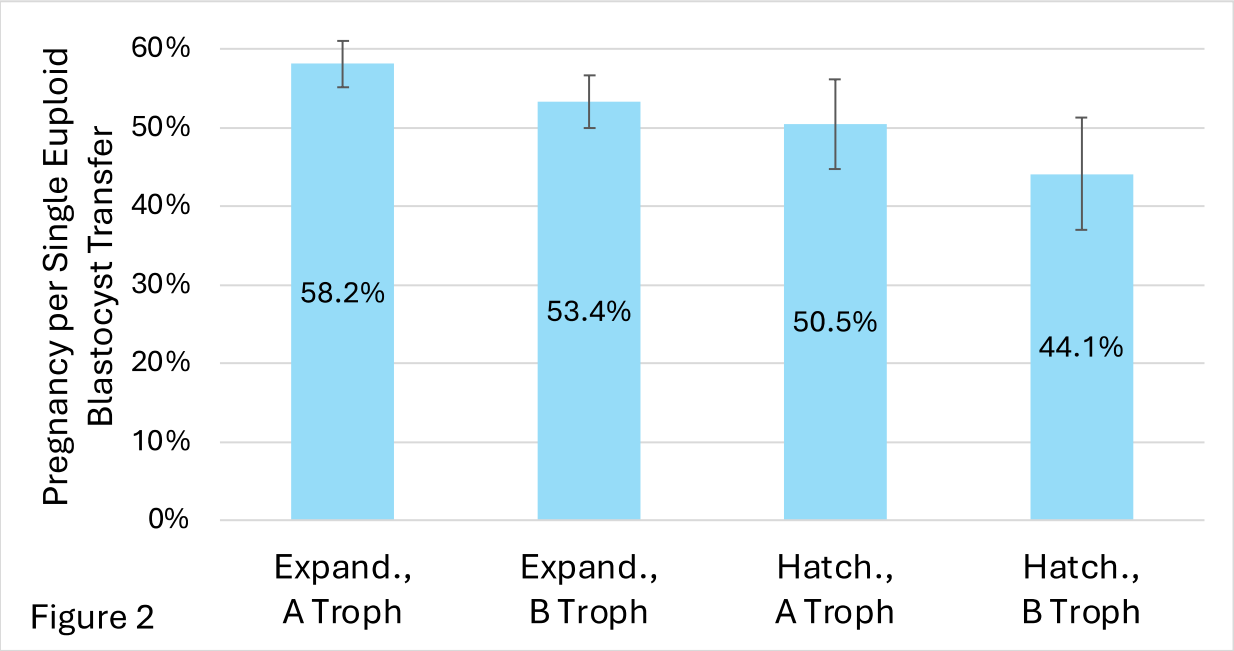

Figure 2

Cryosurvival was usually (88%) excellent. Among the remaining embryos, pregnancy dropped substantially with declining cryosurvival (57% for excellent, 40% for good, 24% for fair, and 0% for poor cryosurvival, respectively, p<.0001).

Conclusions: This analysis of single euploid blastocyst transfers confirms that standard inner cell mass and trophectoderm grades have similar predictive value as reported for transfers of non-biopsied embryos [1,2]. However, pregnancy rates were significantly lower (48% versus 56%, relative decrease = 16%) among spontaneously hatching/hatched blastocysts compared to blastocysts biopsied and cryopreserved prior to hatching, in direct opposition to reports of non-biopsied blastocysts, whether transferred fresh [3] or following cryopreservation [4]. These unanticipated results suggest that performing trophectoderm biopsy after rather than before spontaneous hatching has begun may be more damaging to the embryo and may significantly reduce its viability and implantation potential. Therefore, forgoing planned biopsy after natural hatching has begun may be warranted.

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References:

1. Ahlström A, Westin C, Reismer E, Wikland M, Hardarson T. Trophectoderm morphology: an important parameter for predicting live birth after single blastocyst transfer. Hum Reprod. 2011 Dec;26(12):3289-96.
2. Hill MJ, Richter KS, Heitmann RJ, Graham JR, Tucker MJ, DeCherney AH, Browne PE, Levens ED. Trophectoderm grade predicts outcomes of single-blastocyst transfers. Fertil Steril. 2013 Apr;99(5):1283-1289.e1.
3. Chimote NM, Chimote NN, Nath NM, Mehta BN. Transfer of spontaneously hatching or hatched blastocyst yields better pregnancy rates than expanded blastocyst transfer. J Hum Reprod Sci. 2013 Jul;6(3):183-8.
4. Han EJ, Park JK, Eum JH, Bang S, Kim JW, Lee WS. Spontaneously hatching human blastocyst is associated with high development potential and live birth rate in vitrified-warmed single blastocyst transfer: A retrospective cohort study. Int J Gynaecol Obstet. 2024 Jan;164(1):315-323.