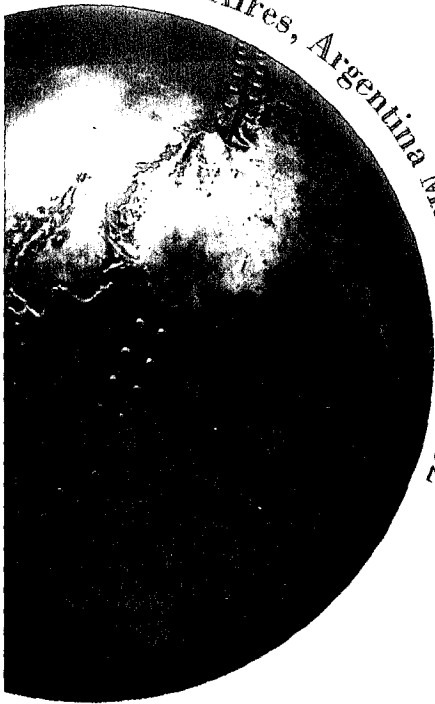


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# **Transferring at Least One Embryo with Eight Blastomeres at 72 Hours Improves Prognosis of Donor Oocyte Recipients Receiving Fresh Embryos but not Frozen-thawed Embryos**

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## **Summary**

The present study evaluated the significance of transferring at least one 8-cell embryo uninfluenced by controlled ovarian hyperstimulation (COH) since, both fresh and frozen embryos were transferred into estrogen-progesterone primed uteri of donor-oocyte recipients.

Significantly higher clinical and viable PRs and implantation rates were found following fresh embryo transfer (ET) but not even a trend was found for frozen ETs.

Thus the adverse effect of COH on uterine receptivity does not seem to explain why fresh 8-cell embryos but not frozen ETs are associated with better outcome following ET. Thus for frozen ETs one should not compensate for embryos with less blastomeres by transferring more embryos.

## **Introduction**

A positive correlation has been observed with the number of 8-cell embryos formed and subsequent blastocyst formation (1). Also clinical pregnancy rates (PRs) and implantation rates were higher in in vitro fertilization (IVF)-embryo transfer (ET) cycles where there was at least one 8-cell embryo transferred on day 3 (2). However, there was less of a difference in PRs and implantation rates when there was at least one 8-cell embryo vs none for frozen-thawed day 3 ETs (2).

One possible explanation for these differences for fresh vs frozen ET may relate to the fact that the former transfer occurred under conditions of controlled ovarian hyperstimulation (COH) whereas the latter transfers were with ovulation suppression and estrogen/progesterone (P) therapy. There are several studies suggesting that COH may adversely affect the uterine environment (3-6). Thus the possibility exists that a more rapidly growing embryo is a heartier embryo and it has a greater capacity to implant into a hostile environment.

To test this hypothesis, the study presented here evaluated whether the presence of at least one 8-cell embryo improves PRs in oocyte recipients where COH is not used.

## **Materials and Methods**

All transfers, fresh or frozen, using exclusively embryos that resulted from fertilization of donor oocytes over a three year period, were evaluated. Clinical PRs, delivery/viable PRs, and implantation rates were determined according to the transfer of at least one eight cell embryo or not. All ETs used three day old embryos.

Recipients without ovarian function are treated with oral micronized estradiol, 2mg x 5 days, 4mg x 4 days, then 6mg x 5 days, beginning on the sixth day of the donor's leuprolide acetate treatment. Recipients with ovarian function are suppressed with leuprolide acetate before starting the estradiol. Recipients are given P vaginal suppositories, 200mg twice daily, and frequently 50mg IM P beginning the day after the donor takes hCG, and transfer occurs on the fourth day of P supplementation.

Generally, twice as many embryos intended for transfer were allowed to cleave to day 3 and the best graded embryos were transferred and the remaining ones were cryopreserved if deemed adequate. Em-

Table 1 - Outcomes of Fresh Cycles According to Blastomere Number

|                      | Maximum cell size <8 | Maximum cell size ≥8 |
|----------------------|----------------------|----------------------|
| # of transfers       | 61                   | 135                  |
| Embryos transferred  | 3.4±1.1              | 3.4±.7               |
| Clinical PR          | 37.7% (23)           | 67.4% (91)           |
| Singleton            | 12                   | 46                   |
| Multiple             | 11                   | 45                   |
| Implantation rate    | 17.2% (36/209)       | 33.9% (154/454)      |
| Spontaneous abortion | 2                    | 14                   |
| Delivery rate        | 34.4% (21/61)        | 57.0% (77/135)       |

bryos were frozen using a simplified freezing technique with a one step thawing protocol (7).

Frozen embryos were hatched as previously described (8). The remaining 2 pronuclear embryos not intended for fresh transfer were also cryopreserved.

## Results

The outcome of fresh ETs is seen in Table 1. Significantly higher clinical PRs, delivery rates, and implantation rates were seen in the group receiving at least one 8-cell embryo.

Interestingly, when evaluating frozen ET, there was no significant difference or even a trend for higher PRs with transfer of an 8-cell embryo versus no 8-cell embryo as seen in Table 2.

The presence of one 8-cell embryo was 68.9% for fresh ETs vs 37% for frozen ETs. Leuprolide acetate was used by 59.8% of recipients with fresh ETs versus only 14.6% of those having frozen ETs.

Table 2 - Outcomes of Frozen Embryo Cycles According to Blastomere Number

|                      | Maximum cell size <8 | Maximum cell size ≥8 |
|----------------------|----------------------|----------------------|
| # of transfers       | 92                   | 54                   |
| Embryos transferred  | 3.4±1.1              | 3.8±1.0              |
| Clinical PR          | 43.5% (40)           | 37.0% (20)           |
| Singleton            | 26                   | 10                   |
| Multiple             | 14                   | 10                   |
| Implantation rate    | 19.4% (61/315)       | 15.7% (31/197)       |
| Spontaneous abortion | 6                    | 4                    |
| Delivery rate        | 37.0% (34)           | 29.6% (16/54)        |

## Discussion

The embryonic genome is fully activated after the 8-cell stage (9). Thus it does not seem likely that an 8-cell embryo is less likely to have a chromosomal anomaly as compared to an embryo with less blastomeres. Thus, the intrinsic capacity of a faster growing embryo to successfully implant does not seem to be because of better genetic selection.

The data presented here show that the discrepancy of higher PRs with 8-cell fresh embryos vs frozen embryos does not appear to be related to a heartier embryo being able to implant better in a hostile environment because 8-cell embryos from oocyte donors resulted in higher PRs and implantation rates even in recipients not receiving COH.

These data, using a different type of patient, reconfirm previous conclusions that embryos with less blastomeres still result in respectable PRs following transfer on day 3 albeit lower than if an 8-cell embryo were present.

Further studies are thus needed to answer the question why blastomere number on day 3 embryos positively correlates with PRs following fresh ET but not frozen ET. From a practical standpoint though one might compensate for the lower implantation rates of embryos with less blastomeres by adding an extra embryo for transfer, such adjustment does not seem necessary for frozen ETs.

## References

1. JONES GM, TROUNSON AO, LOLATGIS N, WOOD C. Factors affecting the success of human blastocyst development and pregnancy following in vitro fertilization and embryo transfer. *Fertil Steril* 70:1022-9, 1998.
2. CHECK JH, WILSON C, SUMMERS-CHASE D, CHOE JK, NAZARI A, LURIE D. Pregnancy rates (PRs) according to embryo cell number at time of embryo transfer (ET). *Clin Exp Obst Gyn* 28:73-7, 2001.
3. PAULSON RJ, SAUER MV, LOBO RA. Embryo implantation after human in vitro fertilization: importance of endometrial receptivity. *Fertil Steril* 53:870-4, 1990.
4. CHECK JH, O'SHAUGHNESSY A, LURIE D, FISHER C, ADELSON HG. Evaluation of the mechanism for higher pregnancy rates in donor oocyte recipients by comparison of fresh with frozen embryo transfer pregnancy rates in a shared oocyte programme. *Hum Reprod* 10:3022-7, 1995.

5. CHECK JH, CHOE JK, KATSOFF D, SUMMERS-CHASE D, WILSON C. Controlled ovarian hyperstimulation adversely affects implantation following in vitro fertilization-embryo transfer. *J Assist Reprod Genet* 16:416-20, 1999.
6. CHECK JH, CHOE JK, NAZARI A, SUMMERS-CHASE D. Ovarian hyperstimulation can reduce uterine receptivity. A case report. *Clin Exp Obst Gyn* 27:89-91, 2000.
7. BAKER AF, CHECK JH, HOURANI CL. Survival and pregnancy rates of pronuclear stage human embryos cryopreserved and thawed using a single step addition and removal of cryoprotectants. *Hum Reprod Update* 2(CD-ROM), 1997.
8. CHECK JH, HOOVER L, NAZARI A, O'SHAUGHNESSY A, SUMMERS D. The effect of assisted hatching on pregnancy rates after frozen embryo transfer. *Fertil Steril* 65:254-7, 1996.
9. BRAUDE P, BOLTON V, MOORE S. Human gene expression first occurs between the 4- and 8-cell stages of preimplantation development. *Nature* 332:459-61, 1988.