Cytoplasmic Transfer in the Human

The Jones Institute Experience
Cytoplasmic Transfer

- In the monkey, Flood et al. (1990) found that the developmental potential of oocytes matured in vitro could be increased by injecting them with the cytoplasm of oocytes matured in vivo.
Cytoplasmic Transfer

- 13% of injected oocytes resulted in pregnancies while none of the sham injected or non-surgical controls resulted in pregnancy. Suggests that factors may be present within the cytoplasm that control genetic, maturational, and/or developmental properties.
Cohen and coworkers (1997) reported the first human pregnancy following the transfer of cytoplasm from donor eggs in the eggs of the recipient.

- The goal of the procedure was to provide healthy cytoplasmic "factors" to the eggs of patients who repeatedly produce embryos of poor quality.
Cytoplasmic Transfer

The Norfolk Experience with Cytoplasmic Transfer

- Patients divided into two groups according to indication for the procedure:
  - Wife is 40 years of age or older, or
  - Couple has had at least two previous IVF attempts which resulted in only poor quality embryos
- Study approved by the IRB of EVMS and allows for the treatment of 15 consenting couples
Patient's eggs may not survive the procedure. Procedure cannot be done that thawed donor eggs may not survive so that risk of obstetric complications unknown if the procedure will increase the been determined. Effect of procedure on eggs or ability of the

RISKS TO PATIENT:

Informed Consent
could adversely affect the offspring
the patient’s eggs and it is unknown if this
may be transferred from the egg donor to
there is a possibility that genetic material
adversely effects the offspring
eggs may be damaged in some way that
abnormalities
risk of obstetric complications or fetal
not known if the procedure will increase the

Risks To Offspring:

Informed Consent
Identified at this time, there may be other risks that cannot be
patients in this study regardless of age.
Aminocentesis is recommended for all
at what rates they would occur.
Way to determine what the exact risks are or
Because the procedure is new, there is no
ALSO STRESSES:

Informed Consent
The pipet, with the tip filled in the tip, is inserted into the donor egg.
Cytoplasm, along with the sperm, is drawn up to the pipet.

Donor Egg
The pipet is moved to the recipient egg and inserted. After aspirating a small amount of cytoplasm, the injected cytoplasm is released along with it.

Recipient Egg
Results

Patients 40 years of age and over:

- 8 patients in 8 cycles
- Average age of 44 (range 41 to 47 yoa)
- Procedure did not appear to have an effect on embryo quality
- No pregnancies were established
Patients 40 years of age and over:

- 39 eggs retrieved (3.2, range 1 to 12)
- 21/39 (54%) 2PN fertilization
- Cytoplasm obtained from 9 donors ages ranging from 25 to 29
- 35/57 (61.4%) survived the thaw
Results

Patients with history of poor quality embryos:

- 3 patients in 3 cycles
- Ages of patients were 35, 35 and 38
- Procedure did appear to have an effect on embryo quality
- One twin ongoing pregnancy was established
Patients with history of poor quality embryos:

- 42 eggs retrieved (mean 14.3, range 7 to 19)
- 26/42 (62%) 2PN fertilization
- Cytoplasm obtained from 3 donors ages 30, 30 and 29
- 27/41 (66%) survived the thaw
Problem: Inadvertent Transfer of Chromosomes
Evaluation of Meiotic Spindle and Chromosomes


- Oocytes evaluated resulted from either clinical or research procedure for cytoplasmic transfer
- 12 oocytes were thawed but not used for transfer and served as controls
- 23 oocytes were thawed and survived the donation procedure as served as tests
Results

- All control oocytes (12/12) demonstrated a normal meiotic spindle apparatus.
- Two of the test oocytes that donated cytoplasm (2/23; 8.7%) demonstrated total dispersion of the chromosomes from the metaphase plate and complete disorganization of the spindles.
Results

- No significant difference between the two groups
- Would this rate of meiotic spindle damage be similar to what is seen in oocytes undergo ICSI?
- Could the damage be reduced with the use of a PolScope?
Spindle View™ Imaging System
Cambridge Research and Instrumentation, Inc.

- Allows for visualization of the spindle during a procedure
- Currently used by laboratories during clinical ICSI cases and research involving enucleation
The Transfer of *In Vivo* Matured Ooplasm into Prophase I Human Oocytes before and after Maturation

- Work performed by Samuel Brown, M.D.
- The developmental failure of human embryos derived from oocytes “matured” in vitro may be due to the deficiency of cytoplasmic factors.
- Would human prophase I oocytes become developmentally competent after microinjecting them with the ooplasm of human MII oocytes matured in vivo?
The Transfer of *In Vivo* Matured Ooplasm into Prophase I Human Oocytes before and after Maturation

- Hypothesized that such an injection would improve fertilization and blastocyst development of these immature eggs.
- If true, salvaging immature oocytes may improve pregnancy success in *in vitro* fertilization patients.
The Transfer of *In Vivo* Matured Ooplasm into Prophase I Human Oocytes before and after Maturation

The effect of cytoplasmic transfer from in vivo matured eggs into PI eggs:

- Control eggs: 14/19 (74%) matured to MII
- Sham eggs: 8/16 (50%) matured to MII
- CT eggs: 11/19 (58%) matured to MII

Suggests that injecting a substance into an egg may have a negative impact on maturation. Results not significant.
The Transfer of *In Vivo* Matured Ooplasm into Prophase I Human Oocytes before and after Maturation

Fertilization results of Control, Sham and Test Groups

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<td>Cytoplasmic Transfer</td>
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The Transfer of *In Vivo* Matured Ooplasm into Prophase I Human Oocytes before and after Maturation

The effect of cytoplasmic transfer from in vivo matured eggs into in vitro matured eggs

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<tr>
<td>Control</td>
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<td>9 (53%)</td>
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<tr>
<td>Cytoplasmic Transfer</td>
<td>17</td>
<td>8 (47%)</td>
<td>5 (29%)</td>
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Conclusions

- Cytoplasmic transfer, if provided clinically, should move forward cautiously and with the full consent of the patients.
- Most patients, having to choose between donor egg and cytoplasmic transfer, would not be bothered with the fact that the child may have genetic material from the mitochondria of the egg donor.