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## TV5 & TV6

The Canadian Fertility and Andrology Society

# Preimplantation Genetic Testing for Monogenetic Disease (PGT-M) with more than one Inherited Diseases in One Family


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## Background and Objective

**Preimplantation genetic testing is offered to the family with monogenetic diseases to avoid affected offspring to be delivered. In most families, PGT-M is performed for one single disorder, and presence of more than one major disorder in a couple is rare. In this study, we report our clinical experience of several PGT-M cases with couples carrying two different genetic diseases.**

## Materials and Methods

**The couples were referred to our clinic for PGT-M.**

****Couple A: 23 years old female patient and 28 years old male patient were first cousins. The Female was diagnosed with Retinoblastoma (autosomal dominant disorder) when she was 5 months old and had surgery to remove her left eye and had radiotherapy for her right eye. The couple's daughter died at 3 and half months of Epidermolysis Bullosa (autosomal recessive disorder). Follow up of the couple revealed that both of them were carriers of this disorder. The female patient was heterozygous in c.241C>T for LAMB3 gene and heterozygous in c.1723C>T for RB1 gene. The male patient was heterozygous in c.241C>T for LAMB3 gene and normal for RB1 gene.**
****Couple B: 32-year-old female and 35-year-old male, the female was affected with DM1 (300 repeats in DMPK gene), and the male was affected with NF1 (c.1381C>T in NF1 gene). They had a DM1-affected son.**
****Couple C: 29-year-old female and 30-year-old male, the female was affected with DM1 (400 repeats) and heterozygous carrier of c.621+1G>T mutation, and the male partner was a heterozygous carrier of delta-508 mutation in CFTR gene. They had one unaffected daughter and had an abortion at 16 weeks following a positive prenatal diagnosis for CF.**

**The study was approved by the research or clinical ethics board of MUHC. The IVF and PGT-M were carried out according to the procedure performed in our center. Embryos diagnosed as unaffected were transferred on day 5 or 6 post-fertilization.**

## Results

**For couple A, 7 full informative and 1 partial informative markers selected from 15 STR markers plus LAMB3 allele were used for the diagnosis of LAMB3. Two full informative and 4 partial informative markers selected from 11 STR markers plus RB1 allele were used for the diagnosis of RB1. For couple B, 7 informative markers selected from 14 STR markers plus DM1 allele were used for the diagnosis of DM1, and 8 informative markers plus NF1 mutation were used for the diagnosis of NF1. For couple C, 7 informative markers plus DM1 allele were used for DM1 diagnosis, and 10 informative markers selected from 17 STR markers plus two specific mutations were used for the diagnosis of CF. Preliminary experiments were performed in genomic DNA and single or few cells, and the overall amplification rate and allele dropout rate (ADO) at cell level were 99% and 4%, respectively, for couple A, 97% and 6%, for couple B, and 98% and 5% for couple C.**

**One IVF and PGT cycle was carried out for couple A. Fifteen embryos were biopsied on day 5 and tested. Five embryos were diagnosed as unaffected for both diseases. Four single embryos were transferred in four separate transfers, leading to two healthy boy delivery. Seven IVF and two PGT cycles were carried out for couple B. Eight embryos were tested but none of them was diagnosed as unaffected for both diseases, resulting in no embryo transfer. For couple C, 5 IVF and 3 PGT cycles were carried out. A total of 16 embryos were tested, and 4 embryos were diagnosed as unaffected for both diseases. Two single embryos were transferred in two separate transfers, leading to one clinical pregnancy, but this pregnancy resulted in a miscarriage at 12 weeks.**

## Conclusions

**The successful IVF-PGT-M cycle mostly depend upon the number of unaffected embryos available for transfer. Our finding shows that it is possible to achieve successful pregnancies but it can also be challenging for some couples who are carriers of more than one inherited genetic condition in a PGT-M setting, especially for female patient with poor ovarian reserve.**

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**Figure 1 Family Pedigree of RB1 for PGD in Family A**

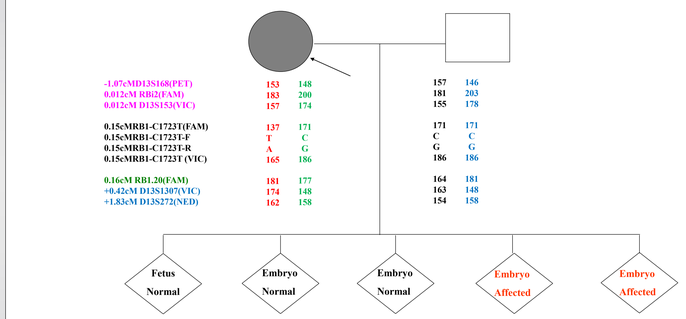


**Figure 2 Family Pedigree of EDB for PGD in Family A**

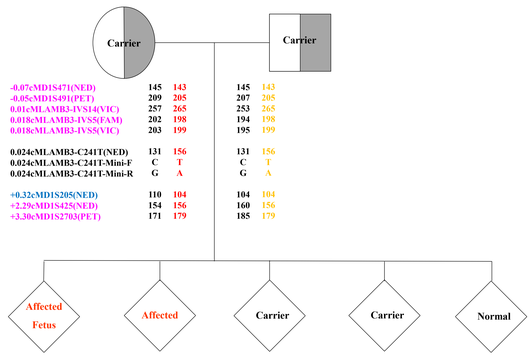


**Table 1 IVF-PGD Cycles for Family 1 (DM1+NF1)**

| PGD Cycle | IVF Cycle | Follicles Retrieved | Follicles Reached MII Stage | Embryos Fertilized | Embryos Biopsied | Normal Embryo for DM1 | Normal Embryo for NF1 | Transferrable Embryo | Embryo Transferred |
|---|---|---|---|---|---|---|---|---|---|
| 1st | 1st | 2 | 2 | 2 | 3 | 0 | 1 | 0 | N/A |
| | 2nd | 3 | 2 | 2 | | | | | |
| | 3rd | 5 | 2 | 2 | | | | | |
| | 4th | 7 | 6 | 5 | | | | | |
| 2nd | 5th | 4 | 3 | 3 | 5 | 3 | 1 | 0 | N/A |
| | 6th | 2 | 2 | 2 | | | | | |
| | 7th | 4 | 3 | 1 | | | | | |

**Table 2 IVF-PGD Cycles for Family 2 (DM1+Cystic Fibrosis)**

| PGD Cycle | IVF Cycle | Follicles Retrieved | Follicles Reached MII Stage | Embryos Fertilized | Embryos Biopsied | Normal Embryo for DM1 | Unaffected Embryo for CF | Transferrable Embryo | Embryo Transferred | Pregnancy |
|---|---|---|---|---|---|---|---|---|---|---|
| 1st | 1st | 15 | 14 | 6 | 5 | 1 | 3 | 1 | 1 | Yes |
| 2nd | 2nd | 7 | 7 | 5 | 5 | 2 | 5 | 2 | 1 | No |
| | 3rd | 13 | 12 | 5 | | | | | | |
| | 4th | 16 | 15 | 11 | | | | | | |
| 3rd | 5th | 13 | 10 | 9 | 6 | 1 | 3 | 0 | N/A | N/A |

# Chromosome Abnormalities Including Mosaicism In Preimplantation Embryos From Karyotypically Normal and Translocation Carriers

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## Introduction

Embryonic aneuploidy is one of the main causes of miscarriage and failure of assisted reproductive technology, especially among patients with advanced maternal age (AMA), recurrent implantation failure (RIF), recurrent miscarriages (RM), male factor infertility, and translocation carriers with older age. Many translocation carriers as well as patients with normal karyotypes trying to conceive without success due to fertility issues will opt for in vitro fertilization (IVF) and preimplantation genetic testing (PGT) to ensure that the implanted embryo is free of any chromosomal abnormalities, which will increase the viability of a successful pregnancy. The main objective of this study is to investigate whether embryos from Robertsonian (Rob) translocation carriers have a higher rate of overall chromosomal abnormalities, including mosaicism, compared to karyotypically normal individuals. Additionally, this study will analyze differences in chromosomal abnormalities between male and female translocation carriers and between different indications of karyotypically normal patients.

## Material and methods

A retrospective study was conducted analyzing the clinical outcomes of patients that underwent IVF PGT cycles in one center. Fifty-five cycles from 26 Rob patients consisting of 31 cycles from 18 male Rob patients and 24 cycles from 8 female Rob patients were analyzed. 361 spare embryos from Rob carriers were tested for mosaicism. Furthermore, 145 cycles from 97 karyotypically normal patients that underwent PGT-A with NGS were analyzed and divided by age group (37 or younger and 38 or older). The remaining 261 cycles from 182 karyotypically normal patients that underwent PGT-A with FISH were analyzed and subsequently divided by age group (37 or younger and 38 or older) as well as by clinical indication (RM, RIF, AMA or other factors of infertility).This study was approved by research ethics board of McGill University Health Center (MUHC).

**Table 1. Male Carrier vs Female Carrier Robertsonian Translocation Carrier Clinical Outcomes**

| | Male Carrier | Female Carrier | Summary |
|---|---|---|---|
| No. cycle (patients) | 31(18) | 24(8) | 55(26) |
| Average female age | 34.8±3.9 | 35.7±4.6 | 35.2±4.2 |
| No. COC (per cycle) | 488 (15.5) | 288 (12) | 776(14.1) |
| 2PN (per cycle) | 236(7.6) | 179(7.5) | 415(7.5) |
| No. of biopsy (per cycle) | 207(6.7) | 174(7.2) | 381(6.9) |
| No. Em tested (per cycle) | 195(6.3) | 168(7) | 363(6.6) |
| Succ tested (%) | 195(100%) | 166(98.8%) | 361(99.4%) |
| Normal (%) | 61(31.2%) | 39(23.5%) | 100(27.7%) |
| Abnormal (%) | 134(68.7%) | 127(76.5%) | 261(72.3%) |
| Cycles with ET | 27 | 19 | 46 |
| No. embryo tranferred (per cycle) | 42(1.56) | 26(1.37) | 68(1.48) |
| No. Sac (IR) | 20 (47.6%)[a] | 4(15.4%)[a] | 24(35.3%) |
| CPR/Cycle | 51.6%[b] | 12.5%[b] | 34.60% |
| CPR/ET cycle | 59.3%[c] | 15.8%[c] | 41.30% |
| Miscarrage (%) | 3/16(18.7%) | 1/3(33.3%) | 4/19(21.1%) |

Abbreviations: No. Em tested = number of embryos tested, Succ tested = embryos that were successfully tested, ET = embryo transfer, No. Sac = number of gestational sacs, IR = implantation rate, CPR = clinical pregnancy rate

a: p= 0.0069, b: p= 0.0025, c: p=0.0032

**Table 2 – Clinical Outcomes of Robertsonian Translocation Carrier Patients vs Karyotypically Normal Patients That Underwent PGT using FISH**

| | Rob carrier total | K.N.37 and younger |
|---|---|---|
| No. cycle (patients) | 55(26) | 92(73) |
| Average female age | 35.2±4.2 | 33.6±3.2 |
| No. COC (per cycle) | 776 (14.1) | 1714 (18.6) |
| 2PN (per cycle) | 415(7.5) | 1091(11.8) |
| No. of biopsy (per cycle) | 381(6.9) | 948(10.3) |
| No. Em tested (per cycle) | 363(6.6) | 867(9.4) |
| Succ tested (%) | 361(99.4%) | 836(96.4%) |
| Normal (%) | 100(27.7%)[a] | 331(39.6%)[a] |
| Abnormal (%) | 261(72.3%) | 505(60.4%) |
| Cycles with ET | 46 | 87 |
| No. embryos transferred (per cycle) | 58(1.5) | 236(2.7) |
| No. Sac (IR) | 24 (35.3%) | 63(26.7%) |
| CPR/cycle | 34.6% | 46.7% |
| CPR/ET cycle | 41.3% | 49.4% |
| Miscarrage (%) | 21% | 16.3% |

a: p= 0.00008

**Table 3. Clinical Outcomes of Karyotypically Normal Patients That Underwent PGT-A using FISH vs NGS**

| | FISH | | NGS | |
|---|---|---|---|---|
| | 37 or younger | 38 or older | 37 or younger | 38 or older |
| No. cycle (patients) | 92(73) | 169(109) | 71(56) | 74(41) |
| Average female age | 33.6+3.2 | 40.7+2.0 | 33.8+2.2 | 40.4+1.51 |
| No. COC (per cycle) | 1714(18.63) | 2654(15.7) | 1041 (14.7) | 1286 (17) |
| 2PN (per cycle) | 1091(11.86) | 1764(10.44) | 650(9.2) | 738(9.9) |
| No. of biopsy (per cycle) | 948 (10.3) | 1570(9.29) | 370(5.22) | 355(4.76) |
| No. Em tested (per cycle) | 867 (9.42) | 1457 (8.62) | 365(5.14) | 355(4.79) |
| Succ tested (%) | 836 (96.4%) | 1404 (96.4%) | 355(97.3%) | 346(97.5%) |
| Normal (%) | 331 (39.6%)[a] | 415 (29.6%) | 205(57%)a | 114(33%) |
| Abnormal (%) | 505 (60.4%) | 989 (70.4%) | 150(43%) | 232(67%) |
| Cycles with ET | 87 | 158 | 34 | 32 |
| No. Em tranferred (per cycle) | 236 (2.71) | 374 (2.37) | 56(1.14) | 35(1.35) |
| No. Sac (IR) | 63 (26.7%)[b] | 78 (20.8%)[c] | 22 (40%)[b] | 17(48%)[c] |
| CPR/ET cycle | 49.4% | 34.8% | 61.7% | 46.8% |
| Miscarrage (%) | 16.2% | 25.5% | 23.5% | 14.2% |

Abbreviations: No. Em tested = number of embryos tested, Succ tested = embryos that were successfully tested, ET = embryo transfer, No. Sac = number of gestational sacs, IR = implantation rate, CPR = clinical pregnancy rate

a: p< 0.00001, b: p= 0.0095, c: p= 0.0019

## Results

Male translocation carriers had significantly better clinical outcomes compared to female translocation carriers (CPR/ET cycle 59.3% vs 15.8%, p= 0.0032) (Table 1). A low mosaicism rate of 10.14% in spare Rob embryos was observed compared to a value previously obtained by the lab of 48.1% in embryos from karyotypically normal patients. Karyotypically normal patients that underwent PGT-A with FISH or NGS had significantly greater percentages of normal embryos in the 37 or younger age group compared to the 38 or older age group (39.6% vs 29.6%, p< 0.00001; 57% vs 33%, p< 0.00001, respectively) (Table 3). The percentage of normal embryos was significantly higher in karyotypically normal patients that underwent PGT-A with FISH than Rob carriers that underwent PGT-SR with FISH (39.6% vs 27.7%, p= 0.00008) (Table 2).

## Conclusion

This study illustrates the increase in chromosomal abnormalities in Rob carriers compared to karyotypically normal patients undergoing PGT. The effect of gender of carrier on clinical outcomes was also observed, with much better clinical results present in male carriers. The effect of declining oocyte competence with increased maternal age was also observed and supported previously published literature. This study also confirms the effectiveness of PGT in both translocation carriers and karyotypically normal patients.

# The efficiency of different WGA (Whole Genome Amplification) technique in the diagnosis of Preimplantation Genetic Testing (PGT)


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## Background and Objective

**Preimplantation genetic testing for single gene defects (PGT-M) allows couples at risk of hereditary disorders to selectively transfer unaffected embryos to the uterus, thereby avoiding the possibility of termination later in gestation. Female patients of advanced maternal age have a higher risk of producing aneuploidy embryos. This can be avoided by preimplantation genetic screening (PGT-A). Due to the limited amount of material available that can be obtained from an embryo, the whole genome amplification is the most important step for preimplantation genetic test for monogenetic disease and chromosome screening. The objective of our study was to investigate the efficiency of two whole genome amplification techniques for preimplantation genetic testing.**

## Materials and Methods

**Two different WGA techniques were used in this study, PCR-based WGA (SurePlex DNA Amplification System) and Multiple Displacement Amplification (MDA). Samples with 4-7 cells were amplified by SurePlex kit (Illumina) or MDA REPLI-Mini kit (Qiagen) followed by one round of multiplex PCR with several STR markers. The control samples were analyzed by standard nested PCR protocol. The PCR products were analyzed on ABI 3130 Genetic Sequencer (ABI, USA). The GeneMapper software was used to analyze the data (ABI, USA). Amplification rate (AR) and allele-drop-out rate (ADO) were calculated to evaluate the efficiency of different amplification methods.**

## Results

**A total of 104 samples were collected and experiments were carried out using 26 STR markers. More than 2700 fragments were analyzed.**

*** Comparison of PCR-based WGA (Sureplex) method and Nested-PCR method**
**The amplification rate of PCR-based WGA was 64.44% which was significantly different from Nested-PCR (97.24%, P-value<0.0001), and the ADO rate (12.50%) did not differ significantly from that of Nested-PCR (4.88%, P-value=0.1524).**

*** Comparison of MDA method and traditional Nested-PCR method.**
**The amplification rate of MDA (98.96%) was not significantly different from Nested-PCR (97.24%, P-value=0.268), and the ADO rate (2.17%) showed no significant difference from that of Nested-PCR (4.88%, P-value=0.4477).**

*** Comparison of two WGA methods: MDA and PCR-based WGA (SurePlex)**
**The amplification rate of MDA (98.96%) was significantly different from PCR-based WGA (64.44%, P-value<0.0001), and the ADO rate (2.17%) was not significantly different from that of PCR-based WGA (12.50%, P-value=0.0669).**

**Comparison of PCR-based WGA (SurePlex) method and Nested-PCR method**

| | SurePlex | Nested-PCR | P-Value |
|---|---|---|---|
| Amplification Rate | 64.44% | 97.24% | <0.0001 |
| ADO Rate | 12.5% | 4.88% | 0.1524 |

**Comparison of MDA method and Nested-PCR method**

| | MDA | Nested-PCR | P-Value |
|---|---|---|---|
| Amplification Rate | 98.96% | 97.24% | 0.268 |
| ADO Rate | 2.17% | 4.88% | 0.4477 |

**Comparison of MDA method and PCR-based WGA (SurePlex)**

| | MDA | SurePlex | P-Value |
|---|---|---|---|
| Amplification Rate | 98.96% | 64.44% | <0.0001 |
| ADO Rate | 2.17% | 12.5% | 0.0669 |

## Conclusions

**As an overall result, MDA amplification rates were similar to Nested-PCR rates, but PCR-based WGA amplification rates were much lower. However, all three methods showed no significant differences in the ADO rates. There are many reasons for which MDA seems to be associated with better results in PGT-M compared to PCR-based WGA. First, MDA results in larger fragments than PCR-based WGA (2-100 kb compared to 0.2-0.8 kb), which leads to better amplification in the second round of Nested-PCR. Second, MDA offers better coverage compared to PCR-based WGA, and shows less amplification bias, which is necessary for the second round of specific amplification. Third, MDA uses Φ29 DNA polymerase, which has a lower error rate and greater processivity compared to *Taq* polymerase, which is used in PCR-based WGA. In addition, there is less DNA template degradation due to continuous denaturation. The non-significant differences in ADO rates are most likely due to the small sample size. Further research with a larger sample size is necessary.**

**Although MDA generates better results in the diagnosis of PGT-M, SurePlex produces more reliable results for PGT-A in the NGS platform (Illumina). The improvement of MDA to the NGS platform for PGT-A requires further optimization.**

# Assessment of semen parameters in transgender women

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## INTRODUCTION

- Literature suggests transgender women may have poorer semen parameters compared to cisgender men when comparing semen samples aimed at cryopreservation and banking
- Gender affirming hormone therapy (GAHT) can have a detrimental impact on sperm quality, however, some studies suggest a higher proportion of sperm abnormalities even among trans individuals who had never initiated GAHT
- A broader understanding of baseline semen characteristics in this population will assist in counselling for fertility preservation and the use of future gametes in assisted reproduction

## AIM

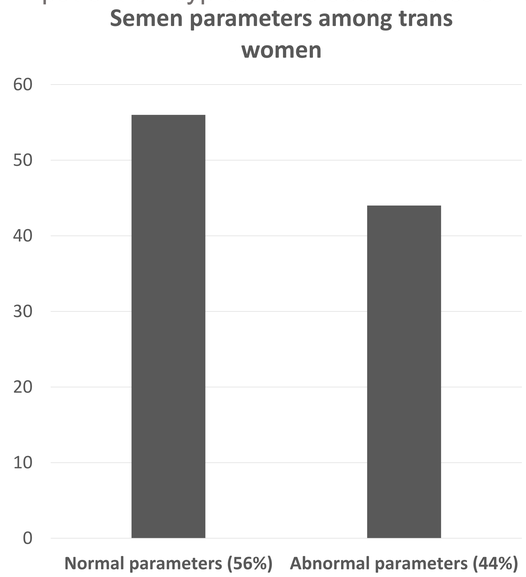
- **Primary Objective:** To evaluate differences in semen parameters of transgender women before initiating gender affirming hormone therapy compared to cis gender, fertile men (WHO Reference Standard). Primary outcomes included semen volume (mL), sperm concentration (M/mL) and sperm progressive motility (%).
- **Secondary Objective:** To compare demographic characteristics between transgender women and cis gender men seeking assisted reproductive technology (ART).
- **Research Question:** Among transfemale patients seeking fertility preservation, are semen parameters reduced compared to cist male fertile controls

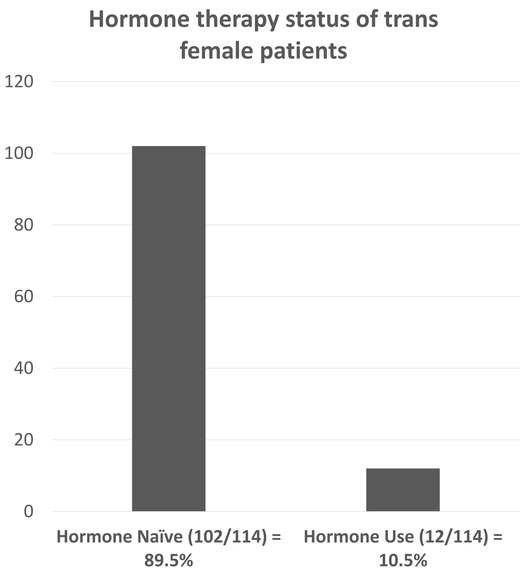
## METHODS

- Retrospective chart review at Ottawa Fertility Centre
- All transgender female patients (≥16 years old) who provided a semen sample for fertility preservation purposes at OFC from 2017 to 2021
- Sperm abnormalities defined as values below the 5th percentile of the WHO reference population of cis males with no infertility
- Analysis of those who never started gender affirming hormone therapy (GAHT) compared to subset of patients who have or were on GAHT
- Exclusion criteria: Any patient who declined enrolment in a research study at OFC
- Demographic data recorded: Age, BMI, social factors (alcohol intake, cigarette smoking, marijuana use, other drugs), pre-freeze semen parameters (volume, concentration, progressive motility), number of straws frozen, post-thaw semen parameters, cryosurvival factor, additional work up (hormones, karyotype, scrotal U/S)

## RESULTS

- 114 unique patients identified during the study period [mean (SD) age 22.6 (5.8)].
  - Mean age did not differ between the groups with normal concentration/motility vs. those below the 5th centile.
- 102 patients had no history of taking hormones prior to sperm banking.
  - 100 patients had sufficient sperm to freeze – pre-freeze volume (mL) 2.8 (1.9), concentration (M/mL) 41.2 (38.4), progressive motility (%) 38.9 (20.9).
- 44/102 (44.1%) patients had either sperm concentration and/or motility below the 5th percentile based on the WHO 6th edition, despite not being on hormone therapy.
- 28.9% of patients in the low concentration or motility group reported use of tobacco and/or marijuana, vs. 19.3% within the group with normal parameters (p=0.26).
- Within the hormone naïve group, 2/102 (2.0%) did not have a sufficient sample to freeze – both azoospermia with normal hormones and normal karyotypes, not investigated further. 11 patients had history of GAHT (Lupron, estradiol, progesterone therapy)
- 3 patients on Lupron were able to freeze after at least 2 months off
- 8 patients on estrogen +/- progesterone were included
  - 2 were unable to freeze sufficient sperm, the remaining 6 did (5 after stopping GAHT)
- 1 patient on cyproterone acetate alone was able to freeze




## DISCUSSION

- It is unknown why semen parameters may have baseline abnormalities within the trans population, however, theories include
  - Increased psychological stress, androgen receptor polymorphisms, underlying genetic disorders and factors specific to the trans population (use of tight undergarments, tucking, under reporting of self-use of GAHT)
  - This theory has been challenged by other literature reporting no differences in gonadotropin levels between trans and cisgender patients, indicating scrotal temperature (and thus specific behaviors) may not be a factor in reduced sperm quality.
- Li et al., 2018 analyzed semen parameters of 141 healthy cisgender sperm bankers and 78 healthy transgender sperm bankers and found that trans sperm bankers had more asthenospermia and worse post thaw parameters. De Nie and colleagues (2020) also concluded that semen quality in transwomen was decreased compared to the general population, and these differences could not be explained by BMI, alcohol consumption, cannabis use, use of GAHT or endocrinopathies.
- Almost half of the patients that chose to pursue fertility preservation at our centre during the study period had semen parameters below the 5th percentile, based on the WHO 6th edition.
- This decrease in parameters pre-freeze increases the likelihood of even lower post-thaw semen quality, which may further necessitate the need for additional investigations which could delay initiation of GAHT and more invasive fertility treatment such as IVF/ICSI to establish a pregnancy in the future.

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# Hemoglobin A1C levels are associated with clinical pregnancy rates in frozen embryo transfers

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## INTRODUCTION

Metabolic dysfunction, including insulin resistance (IR), impaired glucose tolerance (IGT) and hyperglycemia (HG), has been linked to impaired reproductive function[1, 2]. Research shows that women with type 2 diabetes have 64% lower odds of fecundability than women without diabetes[3].

In addition, moderate metabolic dysfunction may also play a role in subfertility. A cohort study in Danish pregnancy planners found that a moderate elevation in HbA1c was associated with reduced fecundability, even when HbA1c levels were within the normal range[4]. Another study in pregnancy planners in Singapore found that increasing plasma glucose levels were associated with time to pregnancy and reduced fecundability, when fasting plasma glucose levels were within the normal range[5]. Mechanisms behind the potential impact of metabolic dysfunction on female fertility are poorly understood. IR, IGT and HG are believed to impact outcomes assisted reproductive technologies (ART)[6,7]. IR in women with PCOS is associated with lower implantation, clinical pregnancy, and ongoing pregnancy rates[8]. Since IR precedes impaired glucose tolerance and is often associated with only mildly elevated HbA1c levels, we evaluated a potential association between HbA1c levels and clinical pregnancy rates in women undergoing frozen embryo transfers.

## METHODS

This retrospective analysis was conducted by identifying all serum HbA1c levels over a period of 1 year in a fertility clinic in Toronto, Ontario. Of those, women who underwent a frozen embryo transfer were identified (65 embryo transfers with 55 women).

The association between HbA1C and clinical pregnancy was evaluated in two ways.

First, Mean HbA1C was compared between participants who achieved, vs. did not achieve a successful clinical pregnancy using a t-test. Second, we compared the proportion of individuals achieving a clinical pregnancy among those with optimal glucose control (HbA1c ⩽5.2%) versus those with suboptimal glycemic regulation (HbA1c >5.2%) using a Chi-square test. Alpha was set at 0.05 and reported p-values are two-sided. Analyses were carried out using Stata (12.1).

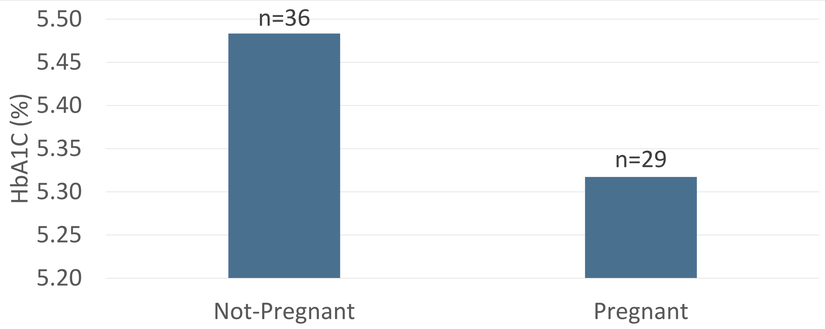
## RESULTS

Sixty-five of participants were recruited from a fertility clinic in Toronto, Ontario, and 29 achieved a clinical pregnancy during the 12mo study period. Participants were 35 years old on average, with mean BMI and peak lining thickness of 25.1 kg/m$^2$ and 9.1mm, respectively. Age and BMI did not differ significantly by either pregnancy or HbA1c status, whereas peak lining thickness was thicker in the group with the highest HbA1c levels. Despite this, there were fewer pregnancies in this group.

**Table 1** – Patient characteristics and pregnancy rates, by HbA1c status

| HbA1C (%) | N-value | BMI (kg/m2) | Age | Peak lining thickness (mm) | Ongoing Pregnancy Rate |
|---|---|---|---|---|---|
| ≤5.2 | 25 | 24.5 | 34.2 | 8.4 | 60.0% |
| >5.2 | 40 | 25.5 | 35.4 | 9.5 | 35.0% |
| P-value | | **0.45** | **0.18** | **0.036** | **0.049** |

**Table 2 –** Mean HbA1C and pregnancy outcomes (p=0.041)




Among participants who achieved a clinical pregnancy, mean HbA1c was significantly lower than in those who did not achieve a clinical pregnancy (5.3% and 5.5%, respectively; p=0.041). Among those with optimal glucose control, 60% achieved a clinical pregnancy, versus 35% of those with suboptimal glucose control (p=0.049).

## DISCUSSION

This exploratory evaluation found an inverse association between HbA1c and clinical pregnancy in women undergoing frozen embryo transfers. Our study had some relevant limitations, including its small sample size, observational nature, and lack of adjustment for potential confounders through statistical analysis. Future work will evaluate a larger sample of individuals achieving euploid embryo pregnancies.

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# It's good to talk: communication between sister cells by cytoplasmic bridges in preimplantation embryos

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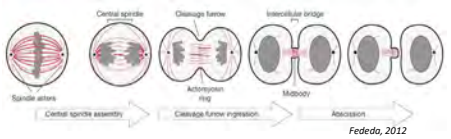
## Introduction

Cytokinesis is the last step of the cell division that physically separates the cytoplasm to form two new cells. Classic experiments demonstrated that large molecules can be shared between some cells within the early embryo. Here we explore the hypothesis that this occurs as a result of a failure of the final step of cytokinesis, called abscission.

Cytokinetic phases in animal cells




Fededa, 2012

Interconnected cells in 8C mouse embryos can share microinjected Horseradish peroxidase.


Lo and Gilula, 1979

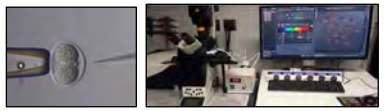
1) Does the cell-cell communication between sister blastomeres in mouse embryos take place through the intercellular bridges that result from failed abscission?
2) Is this inter-cellular communication important for preimplantation development?

## Methods

In vitro mRNA synthesis

**PCNA-EGFP**
(Proliferating cell nuclear antigen-GFP)
**H2B-RFP**
(Histone 2B-RFP)
**PAGFP**
(Photoactivatable green fluorescent protein)

Microinjection of mRNA and Dextran-Fluorescein in 2- and 4-cell embryos and confocal imaging

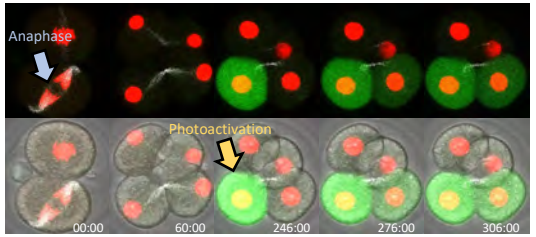


Data analysis

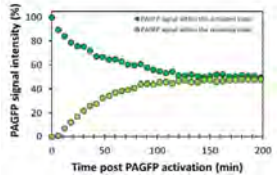
Prism

## Results

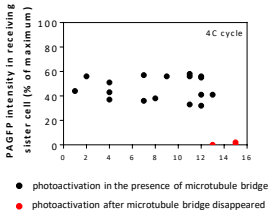
### 1. PAGFP passes from one sister blastomere to another in embryos, and this correlates with the presence of a microtubule bridge.




Photoactivation of Photoactivatable-GFP (PAGFP) in one sister cell 6h after anaphase of the previous division.

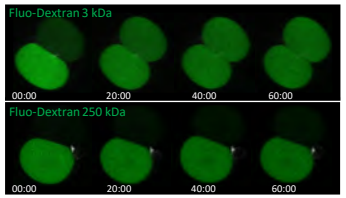


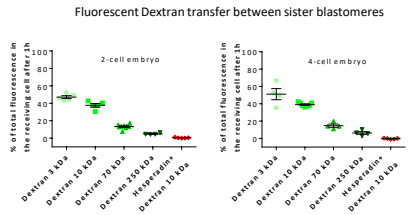

Fluorescent signal of PAGFP equilibrates in both sister cells around 100 min after photoactivation.



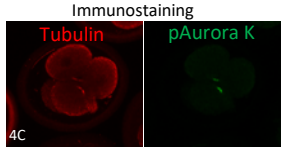

Fluorescent signal of PAGFP detected in prophase in the receiving cell 1h after photoactivation.

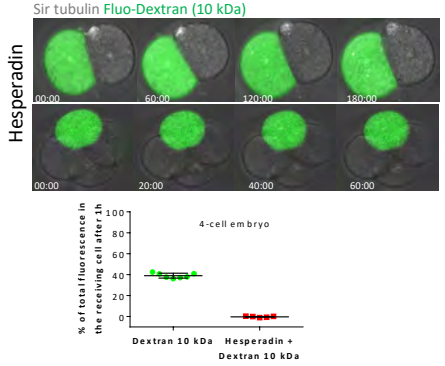
### 2. Molecules in the order of tens of kDa pass through the cytoplasmic bridge.



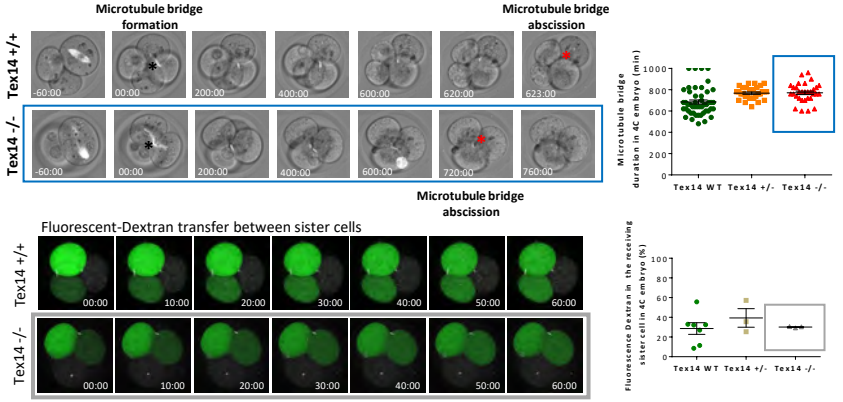




### 3. Aurora kinase inhibition efficiently abolishes both the MT bridge and cytoplasmic sharing of fluorescently-labelled dextran.



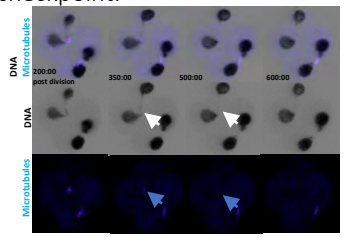



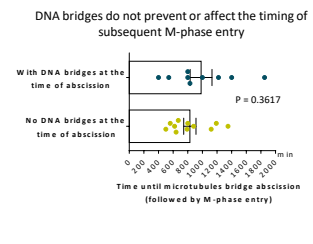

### 4. Sister blastomeres in Tex 14 KO embryos display no changes in microtubule bridge duration, and no loss of cell-cell continuity.




### 5. Abscission failure does not reflect the activation of the DNA-bridge abscission checkpoint.

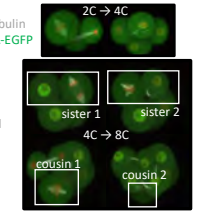
Artificial induction of DNA bridges using Topoisomerase II-α inhibitor - ICRF-193

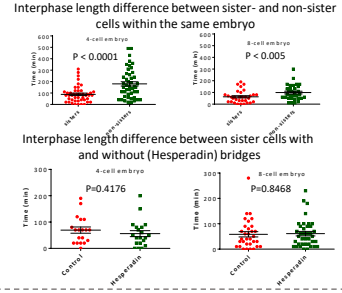


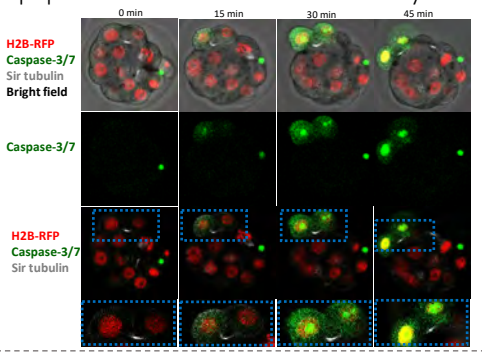

### 6. Sister cells exhibit some cell cycle coordination, but apparently independently of the cytoplasmic bridge.

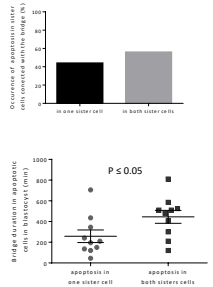
Live confocal imaging from 4C mouse embryo showing the S-phase (PCNA-EGFP) and M-phase onset (Sir tubulin) in the sister- and non-sister cells.




### 7. Apoptosis in sister cells coordinated by the bridge connecting them…?




## Conclusions

- Intercellular bridges in mouse embryos are formed as a result of delayed abscission.
- The intercellular bridge serves as a channel through which cytoplasmic content can be shared, and is dependent upon Aurora Kinase, not on Tex14.
- Mouse embryos lack a DNA-bridge-dependent abscission checkpoint.
- Sister cells in mouse embryos have somewhat synchronized cell cycles, but so far this looks independent of the bridge. Preliminary results suggest a role in coordinated apoptosis.

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# Novel Objective Oocyte Quality Grading Algorithm

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## ABSTRACT

**Introduction:** Standardization is one of the main pillars of any clinical laboratory. With most procedures offered within the scope of IVF lab services standardized by individual teams, oocyte grading happens to be one of the procedures that lacks consistency amongst centers. Currently, each lab performs the assessment of oocytes post egg retrieval per their own criteria where the embryologists record notes on the embryology sheet outlining the inclusions and deviations observed. This current method allows for significant subjectivity based on inter- and intra-procedural deviations amongst embryologists. **The Oocyte Quality Assessment Survey (OQAS) was developed with the aim to standardize oocyte quality assessment.** The survey allows embryologists to grade the cumulus oocyte complexes (COC's), along with oocytes by assigning binary grades through absence/presence of specific morphological inclusions**.** Based on previously published work, these selected inclusions are believed to be the deviating variables of oocyte quality (Wilding M, et al. 2007, Lazzaroni-Tealdi E, et al. 2015).

**Methods:** OQAS was designed by a group of Canadian embryologists based on previously published oocyte grading characteristics. Six Canadian IVF centers participated in the survey with a total of 81 surveys completed. Embryologists assigned binary scores indicating presence or absence of various oocyte morphological criteria during routine oocyte assessments. **Total Oocyte Scores (TOS) were calculated per patient and calculated by totaling the number of inclusions divided by the total number of mature oocytes retrieved**. TOS ratio groups were compared against outcomes of interest. Equal weight was assigned to various oocyte inclusions. Male factor, patients with hormonal disorders, and patients >40y/o were excluded from the study. Data was compared using one-way ANOVA tests.

**Results:** Demographics among the TOS ratio groups were assessed to ensure comparability between groups; a correlation was observed between mean AMH and TOS groups (Table 1). TOS was tested against clinically relevant age groups among fertility patients to confirm defined inclusions were appropriately selected; increased age correlated with increased TOS and trended toward statistically significant (Table 2). Though not statistically significant, trends among total blast conversion and 3BB+ blast conversion were decreasing with increasing TOS leading toward less favourable outcomes (Table 3). Mean total blasts and mean 3BB+ blasts also trended toward significance as higher TOS ratios corresponded with lower mean blast values (Table 4).

**Conclusions:** The newly designed OQAS demonstrates a correlation between diminishing oocyte quality with increasing female reproductive age, as previously published by other groups (Navot D. *et al.,* 1991). A correlating trend of increased TOS was also confirmed to be consistent with decreasing blastocyst conversion rates as well as total number of good quality blastocysts (GQB = 3BB+). Higher TOS generated through increased number of oocyte inclusions within each oocyte cohort translated to diminished embryo development competency when controlling for male factor infertility and female age. Future studies will investigate further relationships.

## BACKGROUND

- Current oocyte quality assessment is subjective and not standardized throughout the embryology labs.
- Standardization and objective assessment is paramount to quality of IVF lab outcomes and patient counseling.
- Objective grading systems also allows for research opportunities in understanding various factors and their effects on oocyte quality.
- Binary grading system built by an Expert Panel of Canadian Embryologists/Lab Directors
- 6 Canadian IVF centers recruiting 81 patients for OQAS participation.

## OBJECTIVE

To create and assess a novel non-subjective oocyte quality measurement tool and understand its correlation with various lab KPI parameters.

## MATERIALS & METHODS

**81 OQAS surveys completed**
**by 6 different Canadian Fertility Centers**

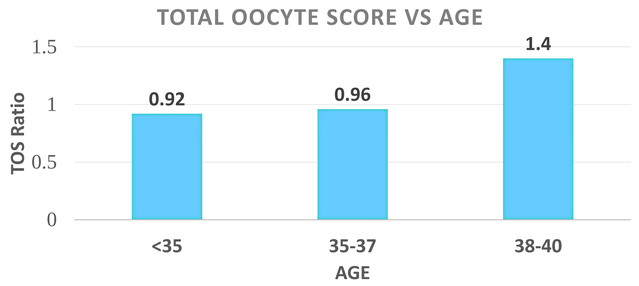
**TOS vs Age** | **TOS vs AMH** | **TOS vs Blastocyst Conversion**

| Oocyte Quality Assessment Survey (OQAS) | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| COC Observations | | | | Oocyte Observations | | | | | | | | |
| | Cumulus Cells (refer to legend) | | | | Oocyte shape | Cytoplasmic Granularity | Vacuoles | Oolema breakage at ICSI | SER | Polar Body | Zona Pellucida | |
| COC Number | Expanded - 0 Compacted - 1 | Light - 0 Dark - 1 | TOTAL SCORE | Oocyte number | Normal - 0 Irregular - 1 | None - 0 Minimal - 1 Extensive - 2 | None - 0 One Vac - 1 Multiple Vac - 2 | Normal - 0 Sudden/difficult- 1 | Absent - 0 Present - 1 | Whole - 0 Fragmented - 1 | Normal - 0 Abnormal - 1 (Thick/Thin/Dark) | TOTAL SCORE |
| 1 | | | | 1 | | | | | | | | |
| 2 | | | | 2 | | | | | | | | |
| 3 | | | | 3 | | | | | | | | |
| 4 | | | | 4 | | | | | | | | |
| 5 | | | | 5 | | | | | | | | |
| 6 | | | | 6 | | | | | | | | |
| 7 | | | | 7 | | | | | | | | |
| 8 | | | | 8 | | | | | | | | |
| 9 | | | | 9 | | | | | | | | |
| 10 | | | | 10 | | | | | | | | |
| 11 | | | | 11 | | | | | | | | |
| 12 | | | | 12 | | | | | | | | |
| 13 | | | | 13 | | | | | | | | |
| 14 | | | | 14 | | | | | | | | |
| 15 | | | | 15 | | | | | | | | |
| 16 | | | | 16 | | | | | | | | |
| 17 | | | | 17 | | | | | | | | |
| 18 | | | | 18 | | | | | | | | |
| 19 | | | | 19 | | | | | | | | |
| 20 | | | | 20 | | | | | | | | |
| Total score | | | | | | | | | | | | |

| IVF Cycle Details | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| IVF cycle attempt # | Number of follicles (1.5+cm at trigger) | # COC's at OPU | # MII | # MI/GV | # Abnormal oocytes (atretic, giant, post-mature) | Fertilization Rate | | Overall Good Quality Blast Rate (≥3BB) | | Total Blast Rate (all grades) |
| | | | | | | % | n/n | % | n/n | n/n |

Figure 1. Oocyte Quality Assessment Survey - binary scores assigned to various inclusions

## RESULTS



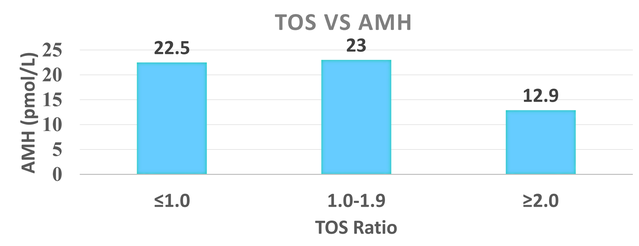




Figure 2. TOS ratio increases with increasing age
Figure 3. TOS ratio increases with reduced AHM
Both figures confirm increasing inclusions with age and reduced AMH

## RESULTS

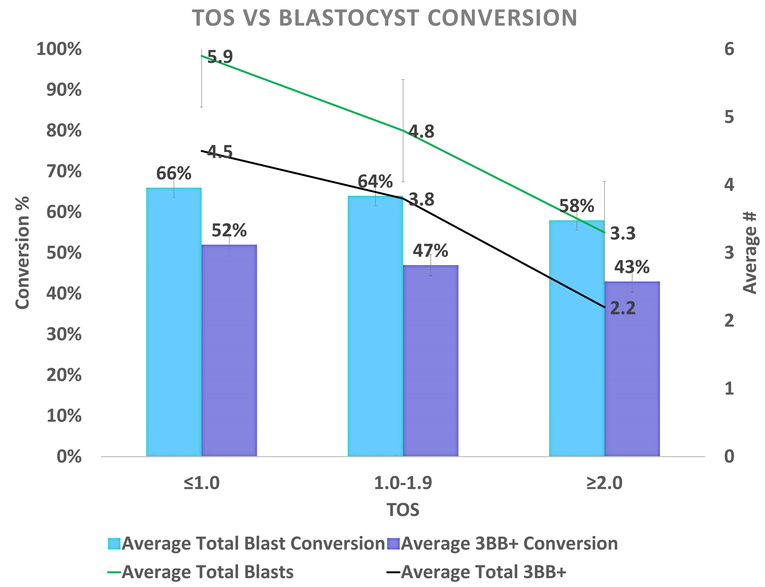



Figure 4. Higher TOS, implying increasing inclusions, correlate with lower blastocyst conversion rates and reduced number of total number of blastocysts.

## CONCLUSIONS

- Newly developed OQAS demonstrates a correlation between diminishing oocyte quality with increasing female reproductive age and decreasing AMH.
- Trend of increased TOS was also consistent with decreasing blastocyst conversion rates as well as total number of good quality blastocysts.
- Increased number of oocyte inclusions results in higher TOS within each oocyte cohort, this translated into diminished embryo development competency when controlling for male factor infertility and female age.
- Future studies to investigate further relationships between oocyte quality through TOS and pregnancy outcomes as well as effects of stimulation protocols and other key clinical laboratory performance indicators on TOS.

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Navot D., Bergh R.A., Williams M.A., Garrisi G.J., Guzman I., Sandler B., Grunfeld L., Poor oocyte quality rather than implantation failure as a cause of age-related decline in female fertility. 1991. *The Lancet.* 337:8754. P. 1375-77.

Wilding M., Di Matteo Loredana, D'Andretti S., Montanaro N., Capobianco C., Dale B., An oocyte score for use in assisted reproduction.2007. *J Assisted Reproduction and Genetics.* 24:350–358.

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# Assessment of the relationship of letrozole stimulated mature follicle number and multiple pregnancy rate (MPR) following intrauterine insemination (IUI)

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## INTRODUCTION

Gonadotropins have been used for ovulation induction (OI) in the setting of subfertility secondary to normogonadotropic anovulation (WHO Class II anovulation) since 1961. Due to the increased MPR in women with polycystic ovarian syndrome (PCOS), the use of clomiphene citrate (CC) and subsequently aromatase inhibitors (AI) are preferred as the first-line treatment(1,2). A recent Cochrane review, updated in 2018, was unable to find a significant difference in MPR between CC and Letrozole (1.7% vs 1.3%; OR 0.69, 95% CI 0.41 to 1.16)(3). Letrozole has been found to have higher Live Birth Rate (LBR) (OR 1.68, 95% CI 1.42 to 1.99) without affecting the miscarriage rate (20% with CC versus 19% with letrozole; OR 0.94, 95% CI 0.70 to 1.26). Furthermore, letrozole has been recommended for the use of OI in the setting of unexplained infertility (4). Yet, little research has been done to further assess the factors influencing multiple pregnancy rate in the setting of letrozole use.

**OBJECTIVE**
- Asses the relationship between the no. of stimulated DF and MPR

**PRIMARY OUTCOME**
- Multiple Pregnancy Rate

**SECONDARY OUTCOMES**
- Clinical Pregnancy Rate, Fetal Sacs, Semen Parameters

## METHODS

**DESIGN** Retrospective Cohort Study
**TIMELINE** Jan. 2013 – Dec. 2018
**DATA COLLECTION** MUHC Reproductive Clinic database (Baby Sentry Management Software)
**ANALYSIS** Statistical analysis was completed with correlation coefficients and multivariate logistic regression. Data is mean ±SD.

| Table 1 | Total |
|---|---|
| **N** | 418 |
| **Age** | 33.6 ± 4.0 |
| **Max ET** | 7.9 ± 1.9 mm |
| **CPR** | 10.5% |
| **MPR** | 0.9% |

*Table 1: Demographics*

**EXCLUSION CRITERIA**
- Age < 18 or > 40 years
- OS with Clomiphene Citrate or Gonadotropins
- Uncharted data

**INCLUSION CRITERIA**
- Age 18 - 40 years
- OS with 5 mg Letrozole in Early Follicular Phase
- Intrauterine Insemination
- Use of hCG trigger

## RESULTS

- Participant's age was 33.6 ± 4.0 years.
- Volume of ejaculate was 2.2 ± 1.5mL. Sperm concentration was 50.5 ± 31.6 mil/mL and motility was 42 ± 16% pre-wash. Sperm concentration 59.7 ± 38.7mil/mL and motility was 85 ± 17% post wash.
- At time of TVUS, the maximum endometrial thickness was 7.9 ± 1.9 mm.
- Clinical pregnancy rate (CPR) was 10.5% whereas MPR was 0.9% overall, and 9% of pregnancies.
- The number of total follicles ≥10mm, DF ± 14 mm or DF ± 16 mm were assessed for a relationship to CPR. No statistical significance was seen with any of the described groups.
- The total number of follicles ≥10mm in mean diameter was unrelated to CP (r= -0.04, p=0.40), number of fetal sacs (FS) (r= -0.32, p=0.51) and number of fetal heart beats (FHB) (r= -0.17, p=0.73).
- For DF ± 14 mm the results for CP, FS, and FHB was (r= -0.009, p=0.86), (r= -0.003, p=0.94) and (r= 0.007, p=0.88) respectively.
- Non-significance was once again seen with DF ± 16 mm CP (r=0.036, p=0.47), FS(r=0.036, p=0.47) and FHB (r=0.054, p=0.27). Multivariate logistic regression analysis was completed to assess predictors of CP. None were identified.

| Total DF | Clinical Pregnancy | No. Fetal Sac | No. Fetal Heart Beats |
|---|---|---|---|
| Total DF > 10 mm | r= -0.04, p=0.40 | r= -0.32, p=0.51 | r= -0.17, p=0.73 |
| Total DF > 14 mm | r= -0.009, p=0.86 | r= -0.003, p=0.94 | r= 0.007, p=0.88 |
| Total DF > 16 mm | r=0.036, p=0.47 | r=0.036, p=0.47 | r=0.054, p=0.27 |

*Table 2: Logistic Regression Analysis*

| *Table 2* | Pre-wash | Post-wash |
|---|---|---|
| **Ejaculate Volume** | 2.2 ±1.5mL | - |
| **Sperm Concentration** | 50.5±38.7 mil/mL | 59.7±38.7 mil/mL |
| **Motility** | 42 ± 16% | 85 ± 17 % |

*Table 3: Semen Parameters*

## CONCLUSION

- We did not show an association between the number of DF, up to 3, and the number of FS or FHB.
- The suggestion is that letrozole stimulated females with 2-3 DF prior to IUI do not require cancellation.
- The data set was too limited to draw conclusions for women with ≥ 4 DF at the time of hCG trigger.
- Further research on the MPR in women with ≥ 4 DF at time of hCG trigger is warranted.

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## DISCLOSURES

The authors have nothing to disclose.

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# Time-lapse KIDScoreD5 and clinical outcomes indicate similar pregnancy potential for multinucleated and non-multinucleated blastocysts

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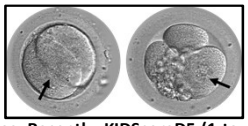
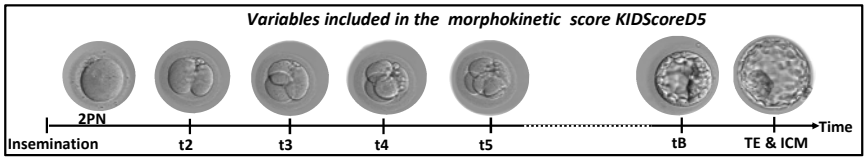
[1] Ottawa Fertility Centre, [2] University of Ottawa, Ottawa, Canada

## INTRODUCTION



Multinucleation (MN) in blastomeres is associated with high rates of chromosomal abnormalities and low rates of clinical pregnancy, especially for cleavage stage embryo transfers (ET). For this reason, many centers prioritize the transfer of non-MN embryos.

Time-lapse embryo ranking algorithms can be utilized to optimize embryo selection strategies. Recently, KIDScoreD5 (1 to 10, 10 being best), a time-lapse multi-variable morphokinetic score, has shown promise as a pregnancy prediction model. Due to all the variables incorporated in the algorithm, KIDScoreD5 surveys the entire embryo preimplantation development, evaluating important developmental milestones and morphological features. Selecting embryos based on highest KIDScoreD5 has the potential of shortening the time to pregnancy. Little is known about the association between KIDScoreD5 result and multinucleation.




*Aim of the study*: To evaluate whether Day 5 MN embryos have the same pregnancy potential as the non-MN ones, and to determine the KIDScoreD5 associated with MN compared to non-MN embryos.

## METHODS

- Retrospective cohort study of consecutive cycles conducted between May 2019 and June 2021.
- Single embryo transfer cycles with either fresh or frozen expanded Day 5 blastocysts.
- Embryo culture in time-lapse incubator EmbryoScope Plus™ (Vitrolife, Sweden).
- Selection for transfer or cryopreservation based on Gardner morphological scoring system.
- Transfers of non-MN blastocysts prioritized over MN.
- The morphokinetic analysis of embryos achieved by assessing the images captured by the EmbryoScope Plus™ (Vitrolife, Sweden), every 10 min in seven focal planes.
- Embryo annotation and computation of the KIDScoreD5™ (Vitrolife, Sweden) performed retrospectively: corresponding to the time of transfer for fresh ET and corresponding to the time of freezing for frozen ET.
- Exclusion criteria: cycles with surgically retrieved sperm, endometrial factors, preimplantation genetic testing.
- Clinical outcomes measured: implantation rate, viable pregnancy rate.
- Statistical test: Chi-square test of independence.

## RESULTS

Table 1. Distribution of cycles based on the multinucleation status of the transferred embryo

| | *Non-MN cycles* | *MN cycles* | *Total cycles* |
|---|---|---|---|
| *Number of fresh single ET (%)* | 223 (83.52 %) | 44 (16.48 %) | 267 |
| *Number of FET (%)* | 329 (81.64 %) | 74 (18.36 %) | 403 |

Figure 1. Distribution of embryos in various KIDScoreD5 categories based on multinucleation status

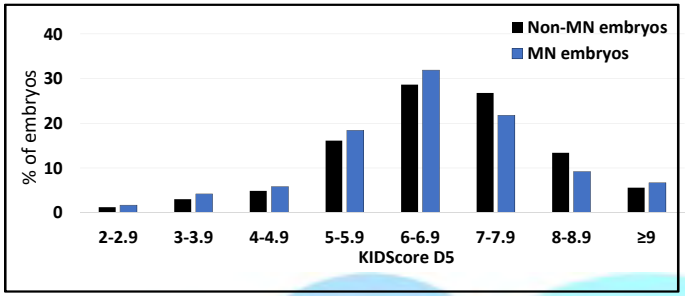
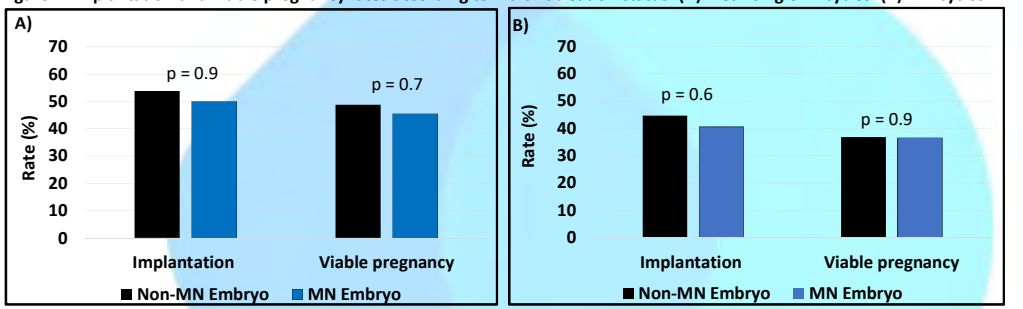



Figure 2. Implantation and viable pregnancy rates according to multinucleation status. (A) Fresh single ET cycles. (B) FET cycles.




## CONCLUSIONS

*DAY 5 BLASTOCYSTS COULD BE SELECTED FOR TRANSFER BASED ON THEIR QUALITY AND MORPHOKINETICS, IRRESPECTIVE OF THEIR MULTINUCLEATION STATUS*

- MN and non-MN blastocysts had a similar distribution of KIDScoreD5, suggesting similar embryo development pattern.
- On Day 5, the multinucleation status of the embryo did not impact the blastocyst ability to implant and result in a viable pregnancy.

# CORRELATION OF MITOCHONDRIAL DNA WITH PLOIDY STATUS AND MATERNAL AGE IN HUMAN BLASTOCYSTS

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## INTRODUCTION

Of all the factors currently available for the evaluation of embryo viability, chromosome status appears to be the most definitive. The current PGT-A methods are capable of accurately determining whether an embryo is euploid, aneuploid, or mosaic. Despite the importance of aneuploidy, this is only one factor amongst many of relevance to embryonic potential, as evidenced by the fact that even the transfer of a euploid embryo can't guarantee a pregnancy. The transfer of mosaic embryos is now considered a possible option for patients undergoing ART with PGT-A testing and in the absence of euploid embryos. Therefore, improved methods for the identification of euploid or mosaic embryos most likely to produce pregnancy would be extremely valuable.

## MATERIALS AND METHODS

This study included 364 blastocysts that underwent PGT-A testing for aneuploidy in our clinic in 2021. It was approved by The Institutional Scientific Advisory Board. The mtDNA ratio was defined as the ratio of mitochondrial DNA to autosomal (chromosome 1-22) DNA. Whole-genome amplification, NGS, and data analysis were performed by Sequence46 using the Ion ReproSeqTM PGS Kit and Ion ReporterTM software (Thermo Fisher Scientific). An ANOVA test was used to compare the variations of the mean mtDNA ratios among different maternal age and different ploidy groups. The percentages of different ploidy embryos were analyzed by using Chi-square test. $P<0.05$ was considered statistically significant.

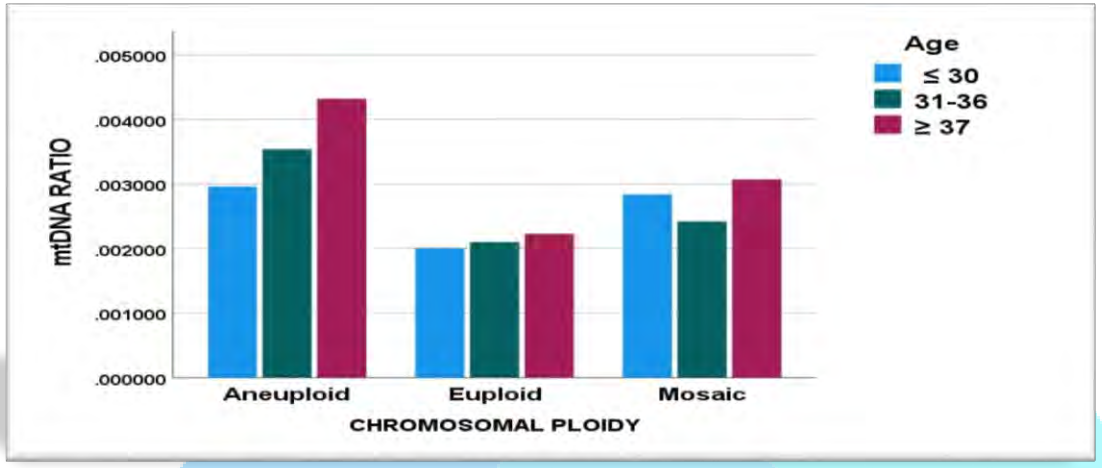
## RESULTS

As shown in Figure 1, the euploid rates of blastocysts significantly decreased with the increase of maternal age while the aneuploid rates of blastocysts were on the contrary. There were no significant differences in mosaic embryo rates among the three different age groups. Regarding the mtDNA ratios, no significant differences were observed among three different age groups in all embryo ploidy statuses: euploidy, mosaic, and aneuploidy. However, significant differences in mtDNA ratios were found among three different ploidy blastocysts (aneuploid > mosaic > euploid) in all three different age groups.

## OBJECTIVES

The assessment of mitochondrial DNA (mtDNA) content as a predictor of embryo viability has recently gained increasing attention in Human IVF. The purpose of this study was to determine whether the mtDNA is associated with embryo ploidy status and maternal age.

**Figure and Table 1: Correlation between mtDNA ratio with ploidy status and maternal age**




| | | Euploid | | Mosaic | | Aneuploid | |
|---|---|---|---|---|---|---|---|
| Patient age | Total # | # (%) | mtDNA | # (%) | mtDNA | # (%) | mtDNA |
| <=30 | 123 | 63 (51%) | 0.00201[a] | 32 (26%) | 0.00284[b] | 28 (23%) | 0.00296[c] |
| 31-36 | 123 | 47 (38%) | 0.00210[a] | 33 (27%) | 0.00242[b] | 43 (35%) | 0.00354[c] |
| >=37 | 118 | 21 (18%) | 0.00223[a] | 25 (21%) | 0.00307[b] | 72 (61%) | 0.00432[c] |

[a,b,c] Values with different superscript letters within the same column or row are significant different ($p<0.05$)

## CONCLUSION

Our results indicate that mtDNA level was associated with ploidy status but not maternal age. The elevated mtDNA level in aneuploid and mosaic embryos may result in more energy needed for chromosomal segregation and active mitochondrial biogenesis at an earlier stage of embryo development, which may cause stress for those embryos to overcome adverse conditions.

# EFFECT OF INDICATION FOR OVUM DONATION ON PREGNANCY RATES IN DONOR EGG CYCLES

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## STUDY QUESTION

***Do patients with previous failed IVF cycles have occult uterine or sperm factors that are not corrected with ovum donation (OD)?***

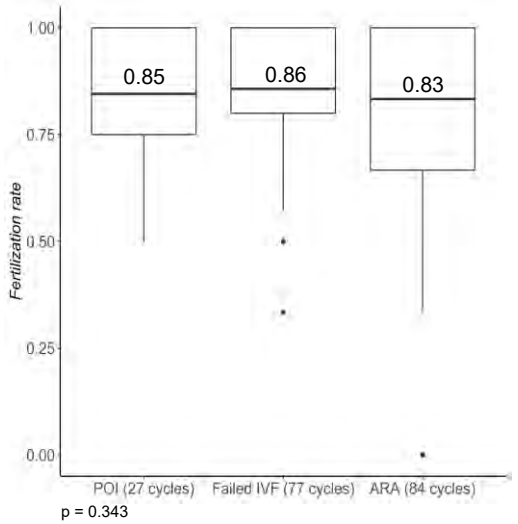
## STUDY DESIGN& METHODS

- Retrospective study of OD cycles February 2019-March 2022.
- Single commercial egg bank.
- G1: premature ovarian insufficiency (POI).
- G2: failed IVF cycles with autologous eggs (<40y).
- G3- advanced reproductive age (ARA) whose initial attempt at fertility was using donor eggs (≥41y).
- Outcomes reported per patient and per cycle.

## RESULTS

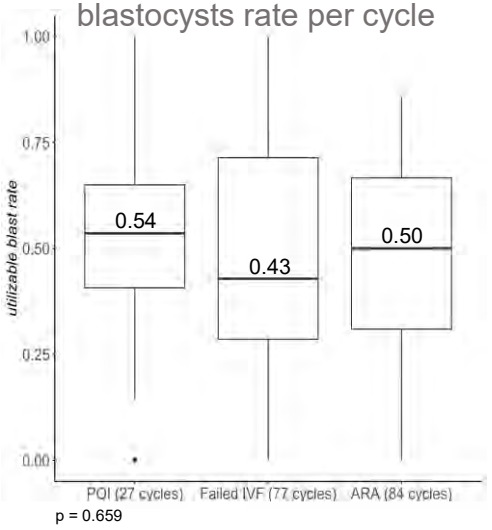
-153 patients (356 cycles) : POI n=22 (35y, ±3.35), Failed IVF n=63 (39y, ±3.08), & ARA n=68 (43y, ±2.45).
-1404 eggs (190 thawed batches), 60.2% fresh embryo transfers (ET) & 17 donor sperm cycles.
-No difference in sperm parameters of pre/post wash with predominately partner (84.4%) & fresh (88.1%) samples.
-No difference in biochemical pregnancy rate (25%, 19%, 15%), clinical pregnancy rate (80%, 77.6%, 78.3%), or spontaneous abortion rate (35%, 24.1%, 31.7%) in G1,G2, and G3 respectively.

Fig.1Fertilization rate per cycle



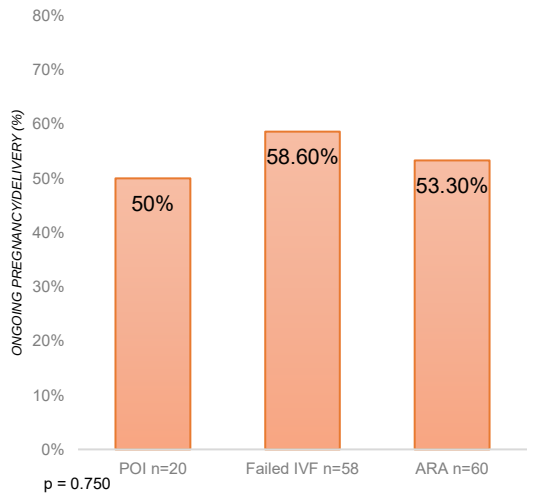

p = 0.343

Fig.2 Utilizable blastocysts rate per cycle



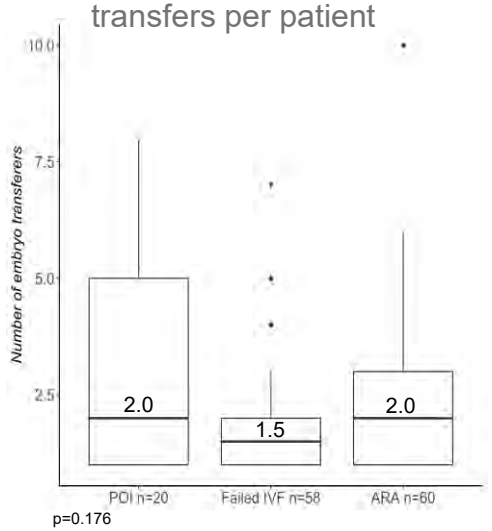

p = 0.659

Fig.3 Ongoing pregnancy & delivery rate per patient




p = 0.750

Fig.4 Total number of embryo transfers per patient




p=0.176

## CONCLUSIONS

- Our results show that infertility in couples with previous failed IVF cycles is corrected with ovum donation, suggesting a minor contribution of uterine or sperm factors to their previous failures.
- Of people who conceived using OD, 75% of those with failed IVF conceived by their 2nd ET while those with POI required 5 attempts.

For more details scan here

SCAN ME