

**RESULTS:** Interpretable results were obtained from 136 embryos. Embryos deemed unsuitable for clinical use displayed an aneuploidy rate of 95.6%, with only 6 embryos demonstrating an apparently euploid chromosomal complement. Of these 6 embryos, 3 were excluded from use due to absence of an inner cell mass and 3 due to inadequate trophoctoderm formation and/or cellular degeneration. Of note, every embryo returning a euploid result underwent cavitation, leaving a 100% aneuploidy rate for embryos that failed to reach the blastocyst stage. When comparing embryos that did or did not attempt to cavitate (excluding triploid embryos), embryos failing to reach this stage contained on average twice the number of chromosomal errors (4.52 vs 2.25,  $p < 0.0001$ ). Unlike previously reported data utilizing clinically viable blastocysts, these embryos exhibited a majority of monosomies over trisomies (65.8% vs 34.2%), likely reflecting the poorer developmental potential of monosomic embryos. Of interest, although triploid embryos are seen at the blastocyst stage, of the 7 triploid embryos identified in this cohort, 6 arrested at either one cell (3 embryos) or following their first cleavage division. The remaining triploid embryo reached cavitation but was of extremely poor quality. This suggests the rate of triploidy in early embryos may be significantly higher than that seen in clinical blastocyst samples. In addition to their standard aneuploidies, 15 embryos contained a segmental aneuploidy, however, in only one embryo was a segmental aneuploidy its only chromosomal error. Similarly, 29 embryos contained additional mosaic aneuploidies, below 50% increase or reduction but distinctly identifiable as a gain or loss.

**CONCLUSIONS:** Embryos failing to reach the blastocyst stage in our culture system displayed universal aneuploidy, a significant finding when counseling our patients. The specific chromosomal errors in non-viable embryos do appear to impact their morphokinetic parameters, however, further work is required to elucidate these potential relationships.

**P-154** Tuesday, October 18, 2016

**INTRACYTOPLASMIC MORPHOLOGICALLY SELECTED SPERM INJECTION (IMSI) DOES NOT IMPROVE CLINICAL OUTCOMES AND EMBRYO PLOIDY IN ASSISTED REPRODUCTIVE CYCLES.** L. Keskinetepe,<sup>a</sup> Z. Beyhan,<sup>b</sup> M. Dayal,<sup>c</sup> J. Hart,<sup>c</sup> M. Keskinetepe.<sup>b</sup> <sup>a</sup>SIRM/Integrated Management, Las Vegas, NV; <sup>b</sup>SIRM, Las Vegas, NV; <sup>c</sup>SIRM, Creve Coeur, MO.

**OBJECTIVE:** It has been suggested that injection of morphologically normal spermatozoa could improve implantation and pregnancy outcomes by promoting better embryo viability, quality and euploidy during IVF-ICSI cycles (1). The intracytoplasmic morphologically selected sperm injection (IMSI) procedure utilizes differential interference contrast microscopy at high magnification (x6600) to select sperm cells without major abnormalities before ICSI (2). Clinical efficacy of the IMSI however, has been controversial and questioned vigorously (3). The objective of the current study is to compare the ploidy status of embryos and the clinical outcomes of PGS cycles randomly assigned to either IMSI or conventional ICSI.

**DESIGN:** Retrospective.

**MATERIALS AND METHODS:** A total of 169 patients were included in this retrospective study, and had undergone PGD/PGS performed as blastomere biopsy for array CGH. The mean ages of the ICSI (Group 1) and IMSI (Group 2) groups were  $35.2 \pm 4.8$  years and  $35.4 \pm 4.3$  years, respectively. Indications for PGS were as follows: advanced maternal age ( $n=29$ ). Male factor ( $n=12$ ), immunologic infertility ( $n=27$ ), decreased ovarian reserve ( $n=32$ ), unexplained ( $n=37$ ), or multiple factors ( $n=32$ ).

**RESULTS:** In group 1, 87 patients had 728 day 3 embryos biopsied, and 33% (240/728) was euploid, 64% (466/728) was aneuploid 4% (29/728) inconclusive. Only 32% of those biopsied embryos on day 3 was developed to blastocyst. In group 2, 82 patients had 723 day 3 embryos biopsied, and 32% (231/723) was euploid, 66% (473/723) was aneuploid 3% (19/723) inconclusive. Only 33% of those biopsied embryos on day 3 was developed to blastocyst. In group 1, 55 patients (63%) had at least one normal blastocyst to transfer on day 5 of the fresh cycle. The PR and IR were 49% (27/55) and 39% (30/77). In group 2, 50 patients (61%) had embryo transfer with at least 1 euploid blastocyst embryos. The PR and IR were 50% (25/50) and 43% (32/75).

**CONCLUSIONS:** Our data suggest that ploidy status of embryos derived by IMSI or ICSI are similar and clinical outcomes of those methods are comparable.

**References:**

1. Bartoov B, Berkovitz A, Eltes F, Kogosowski A, Menezes Y, Barak Y. Real-time fine morphology of motile human sperm cells is associated with IVF-ICSI outcome. *J Androl.* 2002 Jan-Feb;23(1):1-8.

2. Setti AS, Figueira Rde C, Braga DP, Iaconelli A Jr, Borges E Jr. Intracytoplasmic morphologically selected sperm injection benefits for patients with oligoasthenozoospermia according to the 2010 World Health Organization reference values. *Fertil Steril.* 2011 Jun 30;95(8):2711-4.
3. Mauri AL, Petersen CG, Oliveira JB, Massaro FC, Baruffi RL, Franco JG Jr. Comparison of day 2 embryo quality after conventional ICSI versus intracytoplasmic morphologically selected sperm injection (IMSI) using sibling oocytes. *Eur J Obstet Gynecol Reprod Biol.* 2010 May;150(1):42-6.

**P-155** Tuesday, October 18, 2016

**DAY 5, 6, AND 7 BLASTOCYST PLOIDY STATUS STRATIFIED BY PATIENT AGE.** J. Nguyen,<sup>a</sup> R. A. Fields,<sup>b</sup> A. Picou,<sup>c</sup> K. Silverberg,<sup>d</sup> M. VerMilyea.<sup>a</sup> <sup>a</sup>Ovation Fertility, Austin, TX; <sup>b</sup>Fairfax EggBank, Austin, TX; <sup>c</sup>Embryology, Ovation Fertility, Austin, TX; <sup>d</sup>Texas Fertility Center, Austin, TX.

**OBJECTIVE:** To determine if a relationship of ploidy status in day 5, 6, and 7 blastocysts exists amongst patient age groups.

**DESIGN:** Retrospective study in a private in vitro fertilization laboratory.

**MATERIALS AND METHODS:** Blastocysts underwent trophoctoderm biopsy and subsequent comprehensive chromosomal screening over the course of 3 years. PGS results were retrospectively compared to the day of trophoctoderm biopsy and stratified by patient age. Oocytes were fertilized using ICSI and embryos were group cultured to the blastocyst stage for up to 7 days using Continuous Single Culture Medium (Irvine Scientific). On Day 3, all multi-cell embryos were artificially hatched by laser ablation of the zona pellucida. Trophoctoderm biopsy was dependent upon good quality hatching of blastocysts on either Day 5, 6 or 7 that showed a well-defined inner cell mass and trophoctoderm. Biopsied blastocysts were subsequently vitrified for potential future use in a warmed embryo transfer cycle, dependent on a euploid result. Biopsied samples were analyzed by array comparative genomic hybridization (aCGH) or next generation sequencing (NGS) technology by a reference laboratory.

**RESULTS:** 1,915 embryos were biopsied yielding an overall euploid rate of 45% (862). Of the Day 5, 6 and 7 blastocysts, 46% (485), 46% (328) and 35% (49) were determined to be euploid respectively based on the day of biopsy. Chi-square analysis revealed that euploid status is dependent upon day of biopsy ( $p < 0.0001$ ). When stratified by age, a significant difference ( $p < 0.05$ ) in euploid rate exists between D5 & D6 and D5 & D7 blastocysts, with no significance between D6 & D7 blastocysts in patients <34 years old. Blastocysts from patients 35-37 revealed a significant difference ( $p < 0.05$ ) in D5 & D7 and D6 & D7 embryos but no difference between D5 & D6. No significant difference in euploid rate between day of biopsy was shown to exist in the 38-40, 41-42 and 43+ age groups.

**CONCLUSIONS:** Aneuploidy is a very common abnormality in human embryos, particularly for women with advanced maternal age (Franasiak, 2014). Up to 61% of all embryos can be aneuploid in women who are 38-47 years old, although younger women (<35) with good prognosis also have a high rate of aneuploidy (Alfarawati, 2011; Munne, 2006). A recent report also indicates that D6 embryos have the highest euploid rate with D5 and D7 embryos contributing equally to overall ploidy status of an embryo cohort (Vaccari, 2014). Our data suggests that patients <34 have a higher rate of euploid embryos on D5 compared to D6 or D7. Day 7 embryos from patients 35-37 have the highest rate of aneuploidy. Interestingly, day of biopsy does not appear to make a significant difference in the ploidy rate of embryos from patients who are older than 38. Further investigation into the implantation and live birth rates amongst different age groups resulting from the transfer of a euploid embryo(s) biopsied on different days is currently ongoing.

**References:**

1. Franasiak, J.M., et al. The nature of aneuploidy with increasing age of the female partner: a review of 15,169 consecutive trophoctoderm biopsies evaluated with comprehensive chromosomal screening. *Fertility and Sterility* (2014): 656-663.
2. Alfarawati, S., et al. The relationship between blastocyst morphology, chromosomal abnormality, and embryo gender. *Fertility and Sterility* (2011): 520-524.
3. Munné, S. Chromosome abnormalities and their relationship to morphology and development of human embryos. *Reproductive biomedicine online* (2006): 234-253.

4. Vaccari, S., J. Conaghan, and P. Chenette. Extended culture to day 7 increases euploid embryo yield per IVF cycle. *Fertility and Sterility* (2014): e134.

**P-156** Tuesday, October 18, 2016

**BLASTOCYST MORPHOLOGY IS A POOR INDICATOR OF EUPLOID STATUS.** A. Picou,<sup>a</sup> A. Hellmers,<sup>b</sup> K. Silverberg,<sup>c</sup> M. VerMilyea,<sup>b</sup> <sup>a</sup>Embryology, Ovation Fertility, Austin, TX; <sup>b</sup>Ovation Fertility, Austin, TX; <sup>c</sup>Texas Fertility Center, Austin, TX.

**OBJECTIVE:** To evaluate the importance of blastocyst morphology in known euploid blastocysts.

**DESIGN:** Retrospective study in a private assisted reproductive technology program.

**MATERIALS AND METHODS:** The euploid status of blastocysts considered to be of marginal morphology was compared to that of good morphology blastocysts following trophectoderm biopsy and preimplantation genetic screening (PGS). Over a period of 9 months, 683 blastocysts of marginal and good quality were selected for trophectoderm biopsy and aneuploidy screening. All embryos were cultured in Continuous Single Culture Media (Irvine Scientific) to the blastocyst stage. On Day 3 of embryo development, multi-cell embryos were artificially hatched by laser ablation of the zona pellucida. Hatching blastocysts with a differentiated inner cell mass and trophectoderm were biopsied on either Day 5, 6 or 7 of culture. Biopsied embryos were categorized by morphology grade. The embryo grading scheme used in our laboratory is an adaption of the Gardner Grading Scale where AA is best quality and ED is poor quality. Blastocysts that had a grade of CC or better (ie. C or better for both the inner cell mass and trophectoderm) were placed in the good morphology group (Group 1), whereas marginal quality blastocysts (Group 2) were graded as CD, EC, or ED. Biopsied samples were analyzed by comprehensive chromosomal screening (CCS) involving the use of array comparative genomic hybridization (aCGH) or next generation sequencing (NGS) technology by a reference laboratory. Biopsies which resulted in no signal were excluded from the study. All age groups were included.

**RESULTS:** Of the 683 embryos, 554 were categorized into Group 1, while 129 were categorized into Group 2. Group 1 embryos had a normal ploidy rate of 44% compared to 34% for Group 2 blastocysts. Statistical analysis by Chi-Square demonstrated no statistical difference ( $p > .05$ ) between the euploid rate of Group 1 and Group 2 embryos.

**CONCLUSIONS:** Recent studies have suggested that all poor morphology and slower growing blastocysts should be biopsied to determine euploid status and therefore implantation potential (Capalbo, 2014). Our results indicate that marginal quality blastocysts are capable of being euploid at a comparable rate to good quality embryos and should therefore be considered for trophectoderm biopsy and PGS. Further evaluation of the implantation potential of euploid marginal and good quality blastocysts is currently ongoing. Determination of implantation rates of marginal quality euploid blastocysts will take time, as typically the best morphological grade blastocysts are prioritized for transfer. Regardless, these findings suggest that the morphologic criteria used by most IVF laboratories in determining biopsy suitability for blastocysts may need to be reassessed in order to afford patients an optimal opportunity for pregnancy per IVF retrieval.

*References:*

1. Capalbo, A., et al. Correlation between standard blastocyst morphology, euploidy and implantation: an observational study in two centers involving 956 screened blastocysts. *Human Reproduction* (2014).

**P-157** Tuesday, October 18, 2016

**LIVE BIRTHS AFTER TRANSFER OF REBIOPSY AND REVITRIFICATION OF BLASTOCYST THAT HAD "NO DIAGNOSIS" FOLLOWING TROPHECTODERM BIOPSY.** H. Lee, D. H. McCulloh, R. Olivares, A. Goldstein-Tufaro, C. McCaffrey, J. Grifo. OB/GYN, New York University Fertility Center, New York, NY.

**OBJECTIVE:** Report of successful clinical outcomes and live births following transfer of re-biopsied and re-analyzed blastocysts that had yielded no diagnosis from the initial biopsy and array comparative genomic hybridization (aCGH) procedure.

**DESIGN:** Retrospective cohort analysis.

**MATERIALS AND METHODS:** Trophectoderm (TE) biopsy and aCGH for aneuploidy screening was performed on blastocysts generated from 1035 patients between January 2011 to 2014 at an academic based institute. All blastocysts were vitrified immediately following TE biopsy pending aCGH analysis. Forty-six patients who had at least one blastocyst that did not result in aCGH diagnosis opted to undergo warming, re-biopsy and re-vitrification of "no diagnosis" blastocyst in an effort to obtain a diagnosis second time around. Blastocysts resulting in a euploid diagnosis following the re-biopsy procedure were transferred in subsequent frozen embryo transfer (FET) cycles. The main outcome measures were euploidy, implantation and live birth rates

**RESULTS:** Overall, 5060 blastocysts were analyzed from 1035 preimplantation genetic screening (PGS) cycles, 155 (3%) of all biopsied blastocyst had "no diagnosis". Of these 155 blastocysts without diagnosis, 88 blastocysts were warmed, 82(93%) survived and underwent re-biopsy for re-analysis. Twenty-one re-biopsied blastocysts from a total 17 individual patients were diagnosed as euploid (26%) and were suitable for subsequent FET. Seven patients have already undergone a subsequent FET of a re-biopsied single euploid blastocyst. The survival rate for blastocysts undergoing second warming was 100% (7/7) resulting in an implantation rate of 57% (4/7) and birth of three normal healthy babies (male) to date.

**CONCLUSIONS:** A small percentage of TE samples fail to yield a diagnosis following PGS analysis. Our observations show that blastocysts that have undergone PGS with no diagnosis after biopsy can be safely re-biopsied, implant and result in live births despite repeated vitrification, biopsy and warming procedures. This particularly relevant in cases where no other euploid blastocyst are available for transfer.

**P-158** Tuesday, October 18, 2016

**THE EFFECT OF EMBRYO SELECTION BY PREIMPLANTATION GENETIC DIAGNOSIS FOR ANEUPLOIDY (PGD-A) IN PATIENTS WITH ADVANCED MATERNAL AGE.** A. Vereczkey, G. Teglas, L. Nanassy. Versys Clinics Human Reproduction Institute, Budapest, Hungary.

**OBJECTIVE:** The aim of this retrospective analysis is to compare the conventional morphology-based embryo selection (MBS) method to the use of PGD-A in case of patients with advanced maternal age.

**DESIGN:** Retrospective data analysis.

**MATERIALS AND METHODS:** Data were collected between January of 2013 and February of 2016 at our clinic. All autologous cycles were included with advanced maternal age (AMA; 37-42 years of age) and fresh embryo transfer. Patients with known chromosome rearrangement were excluded. In PGD-A group all transferred embryo(s) were known euploid, while in MBS group transferred embryos had an unidentified chromosome profile. Oocytes were fertilized using ICSI or IVF in the MBS group while in PGD-A group only ICSI was used. In case of PGD-A group blastomere biopsies were carried out and the samples were analyzed with 24sure BAC microarrays (Illumina). All embryo transfers were carried out at day 5. Chemical, clinical, ongoing pregnancy and miscarriage rates were recorded. Ongoing pregnancy was noted if pregnancy had completed at least 12 weeks of gestation. Miscarriage was noted in case of pregnancy loss after detected fetal sac.

**RESULTS:** In the PGD-A group 78 euploid embryos were transferred in 63 cycles with an average number of 1.24 embryos per cycle. The chemical, clinical and ongoing pregnancy rates were 50.79%, 44.44% and 36.51%, respectively. In the MBS group the average number of transferred embryos was significantly higher (1.49 vs. 1.24;  $p < 0.01$ ) compared to PGD-A group. No difference was found in chemical (47.06%) and clinical (37.65%) pregnancy rates. However the ongoing pregnancy rate was significantly lower in the MBS group (22.35% vs 36.51%,  $p < 0.05$ ) compared to the PGD-A group. A significantly lower miscarriage (14.29 vs. 40.63,  $p < 0.05$ ) and a higher sustained implantation rates (34.62% vs. 16.54%,  $p < 0.01$ ) were observed in the PGD-A than in the MBS group.

**CONCLUSIONS:** Here we show that PGD-A is a more efficient for AMA patients to select viable embryos for transfer than conventional morphology-based selection. PGD-A not only supported higher ongoing and sustained implantation rates but also lower miscarriage rate. With the utilization of PGD-A not only the number of embryos transferred can be decreased but the risk for miscarriage can be lowered which is a major benefit compare to conventional IVF/ICSI treatments.