PP-046
Relative amount of mitochondria DNA (MTDNA) in euploid and aneuploid blastocysts

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Introduction: Mitochondria play a vital role in preimplantation embryo development where initial stages such as spindle formation, chromatid separation and cell division are supported by mitochondria derived from the oocyte. Elevated levels of mtDNA in human blastocysts were shown to be associated with aneuploidy, advanced maternal age and implantation failure (Fragouli et al, 2015). This is a retrospective study to analyse the relative amount of mtDNA in euploid and aneuploid blastocysts using a single aneuploidy screening platform. This study was performed at Alpha Fertility Centre (AFC), Malaysia from September to December 2015.
Material and Methods: Forty-five patients (age range 23–43) underwent IVF cycles and their embryos were further cultured to blastocyst stage. Blastocysts with at least fair grade (using Gardner blastocyst grading system) were biopsied and screened for chromosome copy number using next generation sequencing (NGS) according to an optimised manufacturer's protocol in AFC (Thermo Fisher, USA). The relative amounts of mtDNA of all euploid (Group A) and aneuploid (Group B) blastocysts were simultaneously assessed using NGS being determined as number of reads for mtDNA/number of reads for genome DNA ratio. The mean age of patients from Group A and B were 33.7 and 35.6 respectively ($p>0.05$).
Results: One hundred and sixteen blastocysts were assessed, of which 60 were euploid (Group A) and 56 were aneuploid (Group B). The relative amount of mtDNA for Group A ranged from 2.0×10^{-4} – 1.4×10^{-3} and the relative amount of mtDNA for Group B ranged from 1.0×10^{-4} – 8.9×10^{-3}. The mean relative amounts of mtDNA for Group A and Group B were 4.0×10^{-4} – 1.0×10^{-3} respectively ($p=0.0061$).
Conclusion: Our preliminary results demonstrated a significantly higher relative amount of mtDNA in aneuploid blastocysts. This supports the findings which indicate increased mtDNA may be correlated to aneuploidy genesis. Since aneuploidy is largely responsible for implantation failure and miscarriage, relative mtDNA might be used as a biomarker to assess implantation potential. However, further validation is required to confirm this proposal.
Keywords: mtDNA, NGS, Euploid, Aneuploid, Blastocyst, IVF

PP-047
Hatching timing and blastocyst quality: Implications on implantation window

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Introduction: It has been widely accepted that the implantation window lasts for 48 hours. However often is not taken into account for blastocyst quality and timing, that blastocysts need to hatch out of the zona pellucida. Healthy trophectoderm may be the reflection of a proper blastocyst molecular factors machinery favoring both hatching and implantation.
Objective: The goal of the present work is to measure the time that cavitated blastocysts require to initiate hatching, whether this timing affects implantation and its association with trophoectoderm quality.
Materials and Methods: Retrospective study from May 2010 until May 2014, including a total of 315 human blastocysts that initiated hatching at the moment of evaluation for embryo transfer; 111 out of 315 had known implantation data. Hatching timings were divided by quartiles; 1st Qt < 109.94 h, 2nd Qt 109.95-114.31; 3rd Qt 114.32-117.31 and 4th Qt >117.32. Statistical analyses of the categorical parameters were done by means of χ^2 test, whereas ANOVA test was utilized for continuous parameters. A p value < 0.05 was considered significant.
Results: From the 315 embryos analyzed, the average time for hatching initiation was 117.39 + 7.38 h. Blastocysts with better trophectoderm quality initiated earlier hatching (112.6 ± 5.5 h) compared to blastocysts with lower trophectoderm quality (113.7±1.2 h), however this difference was not significant. Implantation rates of embryos on each Qt were: 57.1%, 53.3%, 68.8% and 42.9%, for the 1st Qt, 2nd Qt, 3rd Qt and 4th Qt respectively. Although statistically significant differences were not found, there was a clear trend toward having more chances of implantation when the embryo initiated hatching between 114.32-117.31 h.
Conclusions: Both blastocyst quality and precise hatching timings are required to achieve a good synchrony with endometrium. The possible association between the trophoctoderm quality and hatching ability of embryos may be an indirect sign of the competence of blastocyts to get out of the zona to start apposition and invasion.
Keywords: Implantation, Time-lapse, Embryo quality

PP-048
Evaluation of the efficiency of single embryo transfer

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There is a general effort in order to increase the quality of treatment in assisted reproduction, namely eliminate twin and higher order pregnancies. Every laboratory should continuously evaluate their ability to further reduce the number of embryos for transfer while maintaining a reasonable pregnancy rate.
Here, we analyze data collected in the last three years to evaluate the effectiveness of single embryo transfer (SET) in our clinical setting to be able to properly advise patients about their chances before ET.
Data has been retrospectively analyzed between January of 2013 and December of 2015. Clinical pregnancy and implantation rates were compared between patients having fresh ET with either a single (SET) or with two embryos (DET). Cycles where oocyte donation, day 3 ET or preimplantation genetic diagnosis/screening was carried out were excluded.
In our clinic 89.31% of all ETs were carried out on day 5 with an average number of 1.39 embryos transferred between 2013 and 2015. A total of 110 SET and 99 DET were included in the analysis. Patients having SET were older compared to those with two embryos replaced (36.83±0.039 vs. 35.16±0.044, $p<0.01$). However, no difference was found when patients with eSET (having at least one embryo frozen) were compared to DET group (35.50±0.085, $p>0.05$). SET resulted a significantly lower clinical pregnancy (32.73% (36/110) vs. 51.51% (51/99), $p<0.01$) but similar implantation (32.72±0.429 vs. 34.34±0.380, $p>0.05$) rates. Data from elective SETs showed no difference in clinical pregnancy and implantation (47.73% (21/44), $p>0.05$ and 47.73±1.148, $p>0.05$, respectively) compared to DETs. No twin pregnancy was detected in SET group while 27.45% of pregnancies (14/51) were twins in DET group.
It has been showed that when embryo selection is supported with comprehensive chromosome screening SET is just as efficient as DET. In our settings, we did not find any difference in the efficiency of elective SET compare to DET even when the chromosomal constitution of embryos is not known. Not surprisingly a lower pregnancy rate was observed in patients when only a single embryo was available for ET. Based on our results there is no reason to transfer more than one embryo.
Keywords: Elective single embryo transfer, Double embryo transfer, Clinical pregnancy rate, Implantation rate