

The Effects of Laser Manipulation on Biopsy Karyotype in PGS

Introduction

Universally applied preimplantation genetic screening (PGS) protocols for blastocysts rely on the isolation of a trophectoderm (TE) biopsy, with an expectation that the biopsied TE karyotype is representative of the entire embryo. It is imperative for the biopsy collection method not to introduce genetic artifacts in the separated cells, as this could lead to failed tests or false positive/negative interpretations. Current blastocyst biopsy procedures routinely include the use of lasers of infrared or near infrared wavelengths to aid the separation of a cohort of TE cells from the embryo mass. Laser pulses are aimed at interfaces of neighboring cells that are mechanically being pulled apart through suction forces and the instant degradation of cell junctions permits the group of targeted TE cells to fully separate and be recovered for chromosomal analysis. It has been reported that infrared lasers have little to no impact in embryo development and downstream potential. However, recent studies in the fields of dermatology and oncology have shown that infrared and near infrared wavelengths can induce DNA degradation in human cells.

Hence, we set out to determine whether laser intensity and pulse number during TE biopsy affects PGS results.

Materials and methods

Aneuploid blastocysts as determined by PGS (Illumina's Veriseq NGS platform) were used for re-analysis. TE biopsies were collected using methods standard to the industry. The Laser (Hamilton Thorne Lykos v5.12) intensity varied between 400-850 μ s and the number of pulses varied between 4-10 pulses. The biopsies were subsequently processed for PGS by NGS.

Results

All TE pieces analyzed to date (n=12) yielded NGS results of similar quality regardless of laser strength and pulse number used at biopsy collection. All parameters used to assess NGS data quality (resulting genetic profile, DLR, read number pre- and post bioinformatic filtering) suggested insignificant DNA degradation across our tested samples.

Conclusions

From our current data we infer that laser strength and pulse number has negligible impact on PGS results due to degraded DNA. In instances of difficult biopsy isolation, stronger laser strength or repeat pulses may be used without compromising downstream NGS data. The impact of laser intensity and pulse number on subsequent embryo viability is yet unknown.