American Society for Reproductive Medicine 2018 Scientific Congress & Expo
October 6 to 10, 2018 ● Denver, Colorado, USA

Title:

UNDERSTANDING THE SCOPE AND SIGNIFICANCE OF MOSAICISM IN HUMAN PREIMPLANTATION EMBRYOS

Authors:

Sekhon L, Ellis E, Maravilla J, Lee JA, Briton-Jones C, Schadt E, Sebra R, Copperman AB.

Affiliations:

1. Reproductive Medicine Associates of New York, 635 Madison Ave 10th Floor New York, New York, United States, 10022

2. Obstetrics, Gynecology and Reproductive Science, Icahn School of Medicine at Mount Sinai, Klingenstein Pavilion 1176 Fifth Avenue 9th Floor New York, New York, United States, 10029

3. Department of Genetics and Genomic Sciences, Icahn School of Medicine at Mount Sinai, 1425 Madison Ave, New York, NY 10029

4. Sema4, 333 Ludlow Street, South Tower, 3rd floor, Stamford, CT, United States, 06902

Objective:

Mitotic errors are known to occur during post-zygotic cell division, and appear to contribute to aneuploidy in human embryos. Little is known, however, about the genesis of these errors or their biological consequences. Research suggests a higher incidence of mosaicism in biopsied blastomeres from day 3 embryos, compared with the trophectoderm cells of a blastocyst, suggesting that embryos with complex mitotic-derived aneuploidy are more likely to arrest (McCoy et al., 2015). In the past, mosaicism in cleavage stage embryos may have been overestimated due to technical limitations associated with the technology used (i.e. FISH) and single-blastomere analysis. Few studies have compared the incidence of mosaicism across the major stages of embryo development, using NGS- an affordable and accurate method of single-cell analysis of copy number variants (CNVs). The purpose of the study was to utilize modern molecular techniques to characterize the degree of mosaicism according to embryo developmental stage, day of development, viability, and morphology.

Design:

Prospective cohort study on human embryos donated for research.

Materials and Methods:

The study included patients who donated fresh embryos during an IVF cycle between January, 2016 and June, 2016. Embryos underwent routine evaluation from day 3 to 7 of extended culture. Embryos were biopsied, and approximately 4 cells were removed for aneuploidy screening by next generation sequencing (NGS) using the ReproSeq assay to assess copy number variants (CNV). Samples with less than 100,000 reads mapping to hg19 and samples with a Median of the Absolute values of all Pairwise Differences (MAPD) higher than 0.3 were considered to yield poor quality data and were excluded from analysis. Mosaicism was identified based on bioinformatical interpretation of intermediate position chromosome CN falling between disomic and aneuploid thresholds (between 1.2-1.8 or 2.2-2.8). Linear regression was used for analysis.

Results:

Of the total of 78 embryos (41 blastocyst/9 morulae/29 cleavage) that underwent aneuploidy screening, 63 were aneuploid, with 92% (n=58) displaying varying degrees of mosaicism. Blastocyst, morula, and cleavage stage embryos had similar mean numbers of mosaic calls per embryo (6.2 ± 5.4 vs. 8.1 ± 4.3 vs. 7.0 ± 4.5 (p=0.6), respectively). Mosaicism was most often noted to be absent in blastocysts (26.8% vs. 5.2%, p=0.03). The degree of mosaicism was not affected by whether an embryo was viable or developmentally arrested (□=-1.6, p=0.2), the number of cells at the cleavage stage (□=0.4, p=0.2), or ICM grade (□=-0.3 p=0.6). Controlling for day of embryo development, the degree of mosaicism was significantly increased in blastocysts with poor expansion (□=7.3, p=0.001) and low trophectoderm grade (□=-7.9, p=0.001).

Conclusion:

Given the consistent level of mosaicism identified throughout cell division from cleavage to blastocyst stage and the lack of association between mosaicism and developmental arrest, our findings refute the hypothesis that embryos with complex mitotic errors undergo negative selection. It should be noted, however, the majority of cases in which no mosaic calls were made involved blastocysts. The overall prevalence of complex mitotic errors among the embryos studied was significantly higher than that expected for good quality blastocysts; an effect likely due to the inclusion of developmentally arrested and poor morphology embryos. This is the first report of an association between increasing degree of mosaicism and poor expansion and trophectoderm grade. This finding supports the theory of progressive clonal depletion as a compensatory mechanism against postzygotic mitotic errors, which could translate into limited

proliferation and increased apoptosis within the trophectoderm. Future research is required to: 1) overcome technical and biological limitations to the accurate assessment of mosaicism, 2) understand the diverse molecular mechanisms contributing to mitotic error, and 3) to define the impact of mitotic errors in preimplantation embryogenesis.

References:

1. McCoy RC, Demko ZP, Ryan A, Banjevic M, Hill M, Sigurjonsson S. Evidence of selection against complex mitotic-origin aneuploidy during preimplantation development. PLoS Genetics 2015;11:e1005601.