MICROFLUIDIC SPERM SORTING VS. DENSITY GRADIENT: A PRELIMINARY ANALYSIS EXAMINING EMBRYO GRADE AND PREGNANCY OUTCOMES IN IVF-ICSI CYCLES USING VARYING SPERM SELECTION METHODS

Zoran J. Pavlovic[1], Megan Sax[2], Alex Tawa[3], Lauren Grimm[4], Emma Radley[4], Nepheli Raptis[4], Hannah Mandell[4], Christy Oso MA[4], Angie Beltsos MD[4], Roohi Jeelani MD[4],
Affiliations: [1]Rush University Medical Center Department of Obstetrics & Gynecology, Chicago, IL 60661, University of Cincinnati, Department of Obstetrics & Gynecology, Cincinnati, OH, USA, [3]Midwestern University Chicago College of Osteopathic Medicine, [4]Vios Fertility Institute, Chicago, IL 60622

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Introduction

- Male factor infertility is estimated to be the sole cause of infertility in nearly 20% of couples, as well as up to 30 to 40% of couples with infertility attributed to combined male and female factors.[1]
- In the presence of normal semen parameters, male factor etiology may be overlooked, however multiple tests to evaluate sperm quality via sperm DNA fragmentation allow for a more comprehensive analysis for ongoing unexplained infertility.[2]
- High SDF, which some studies have suggested to be >27% fragmentation, may not only reduce pregnancy rate, but also compromise embryo development and increase the risk of miscarriage.[3] SDF may be measured by the sperm chromatin structure assay (SCSA), sperm chromatin dispersion test (SCD), and the terminal deoxynucleotidyl transferase deoxyuridine triphosphate (dUTP) nick end labeling (TUNEL) which can help determine sperm quality.[4,5]
- Microfluidic sperm sorting methods may allow for better selection of healthy sperm from damaged or dead sperm and sperm with less DNA fragmentation and reactive oxygen species.[6]
- Microfluidic sperm sorting is a less traumatic sperm selection method that has been shown to decrease DNA fragmentation and oxidative stress adducts that can potentially improve pregnancy rate and clinical outcomes for couples undergoing assisted reproductive technologies such as in-vitro fertilization with intracytoplasmic sperm injection.[7]

Objective

- To compare embryo quality and pregnancy outcomes, including clinical and ongoing pregnancy rate, between ICSI-IVF cycles that utilized density gradient centrifugation vs. microfluidically selected sperm.

Design

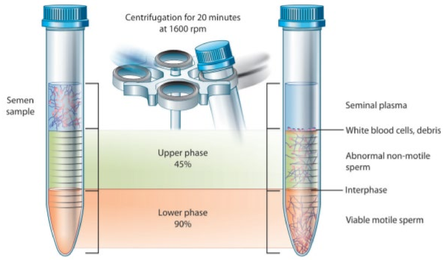
- Retrospective chart review at a private infertility center.

Materials & Methods

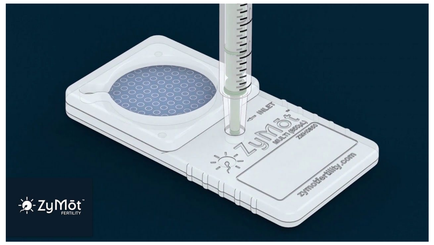
- All patients who underwent in-vitro fertilization (IVF) intracytoplasmic sperm injection (ICSI) cycles via density gradient followed by microfluidic sperm sorting from 2017 to 2019 were included.
- Baseline characteristics included paternal and maternal age, BMI, primary infertility diagnosis and semen analysis.
- Primary outcome: Embryo grade using the Gardner scale that was organized into high (3-6 AB or AA), medium (2-6 BB or BA) and low grade (any embryo with an Expansion Grade of 1 as well as 2-6 BC, CB or CC) groups. Secondary outcome: Clinical pregnancy rate.
- Only embryos that developed to day 4 of incubation and were classified as arrested/morula or given an embryo grade were analyzed. All embryos that did not incubate past day 3 were excluded from analysis.
- Chi-square test of association was used to analyze the data using SPSS (SPSS Inc., Chicago, IL, USA).

Results

- Baseline characteristics of the oocyte source, including age, BMI, and AMH were similar (p>0.05).
- A total of 373 and 376 MII oocytes underwent ICSI with microfluidic and density gradient selected sperm, respectively.
- 73.5% (274/373) of microfluidic IVF-ICSI embryos and 74.2% (279/376) of density gradient IVF-ICSI embryos developed to day 4 of incubation before being classified as either arrested/morula or receiving a grade.
- There was no significant difference in the quantity of embryos that developed beyond a day 4 morula/arrested classification and achieved blastocyst status between the two groups.
- No significant difference was found between the two groups in terms of the number of embryos that were cryopreserved, transferred or discarded.
- When comparing embryo quality there was no significant difference between the number of blastocysts rated as high medium or low quality embryos.
- Finally, out of the embryos that were transferred, only 23% (3/13) in the microfluidic group and 7.1% (1/14) in the density gradient group achieved a clinical pregnancy which was not a statistically significant difference.



Density gradient centrifugation consists of carefully layering upper and lower gradients in a test tube followed by the ejaculate layered on top. The sample is then centrifuged at 1600 rpm for 20 minutes. Clear seminal plasma is retained on the uppermost part of the gradient whereas the immature, abnormal sperm are seen along the gradient based on their density and motility. Highly motile normal sperm move actively to the bottom of the gradient and collected as a pellet.



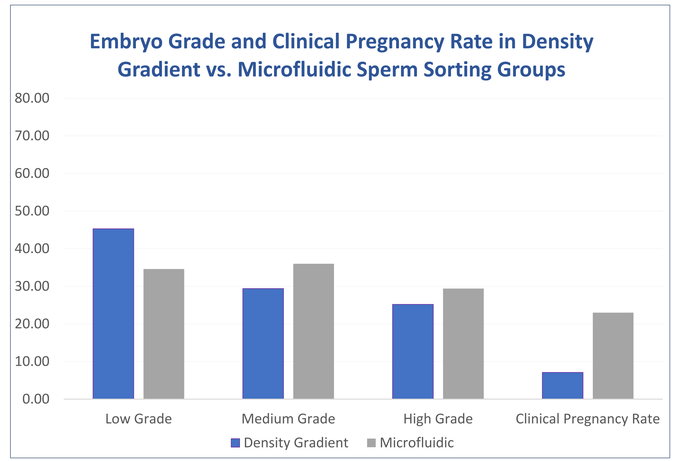
Microfluidic sperm sorting consists of taking 850µL of semen ejaculate and injecting the sample through an inlet port. The semen travels through an outlet port to a section underneath the membrane which consists of numerous 8-micron pores. Fertilization media is placed on top of the membrane. During incubation, the most morphologically normal, motile and genomically competent sperm migrate through the pores and are collected from above.

Embryo Grade and Clinical Pregnancy Rate in Density Gradient vs. Microfluidic Sperm Sorting Groups

	Density Gradient	Microfluidic	p-value
Low Grade	45.3%	34.6%	0.07
Medium Grade	29.4%	36.0%	0.25
High Grade	25.2%	29.4%	0.43
Clinical Pregnancy Rate	1/14 (7.1%)	3/13 (23%)	0.24

Table 2 IVF-ICSI Outcomes

Results



Discussion

- Our results show that there is no significant difference in embryo quality or clinical pregnancy rate between microfluidic sperm sorting IVF-ICSI vs. density gradient selected IVF-ICSI cycles.
- Although microfluidic sperm sorting is thought to cause less oxidative stress/be less traumatic to sperm, similar clinical outcomes in IVF-ICSI can be seen with both density gradient and microfluidics.
- Patients can therefore be confident that the standard of care density gradient method will result in similar outcomes compared to microfluidic sperm sorting at least for IVF-ICSI.
- Further investigation to determine the clinical utility of microfluidic sperm sorting within other treatments such as intrauterine insemination (IUI) is warranted.

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