



**FERTILITY**<sup>®</sup>  
MEDICAL GROUP

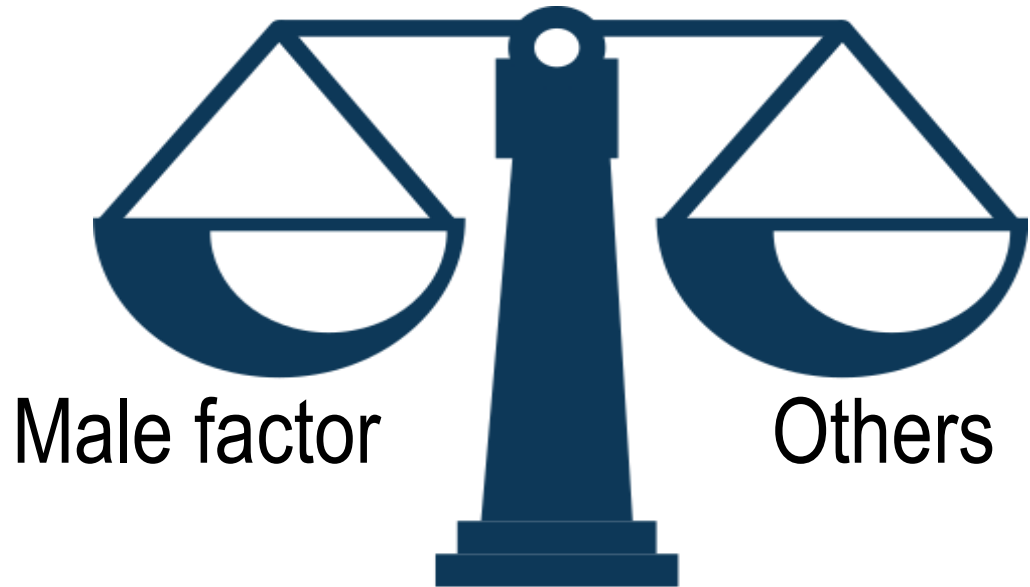
# **Oocyte ability to repair sperm DNA fragmentation: The effect of maternal age on ICSI outcomes**

Edson Borges Jr., Daniela Paes de Almeida Ferreira Braga, Amanda Setti, Rodrigo Rosa Provenza, Assumpto Iaconelli Jr.

# INTRODUCTION



Infertility: 15% of the couples

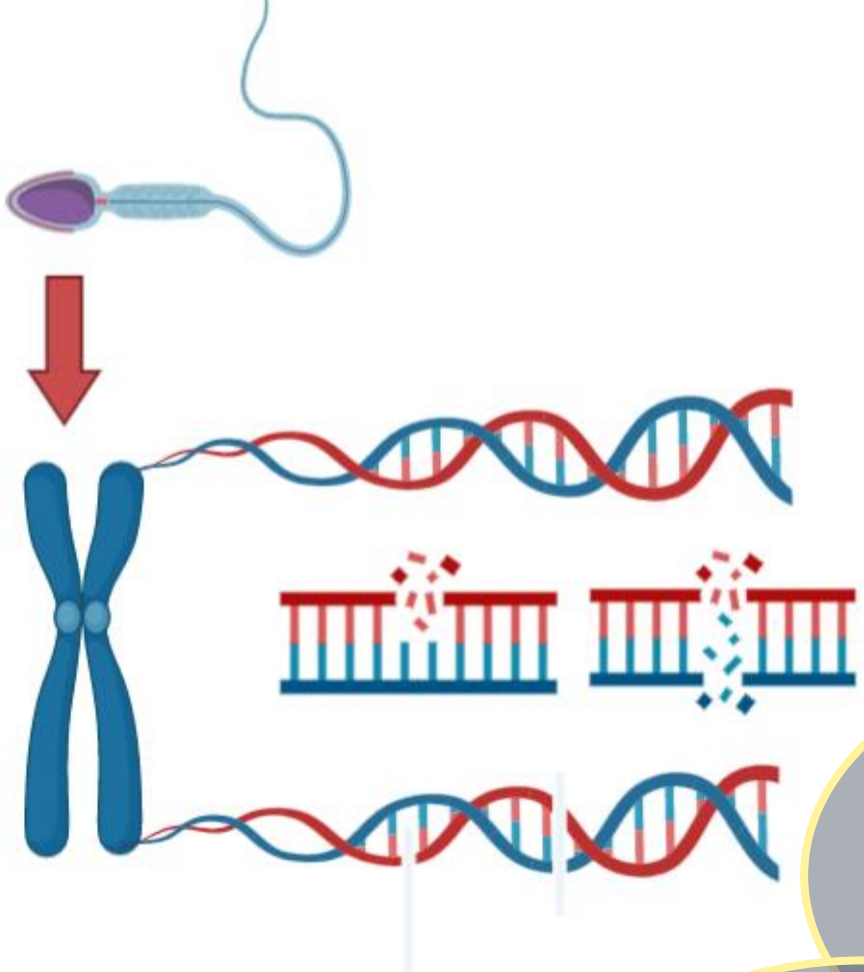
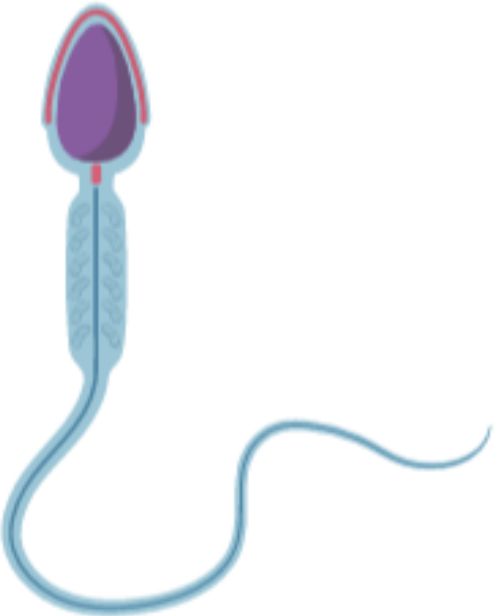


- Sperm Volume
- Sperm count
- Sperm motility
- Sperm morphology

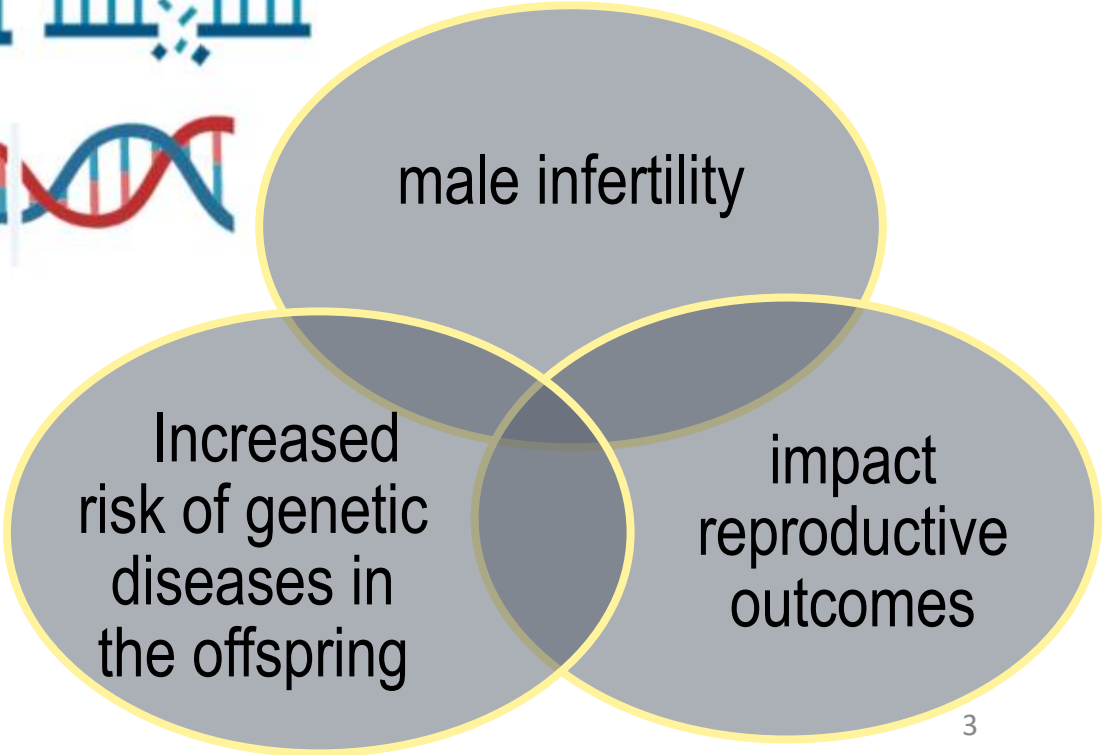
Normal

Abnormal

# INTRODUCTION



Novel methods have been introduced to improve semen analysis at a functional level

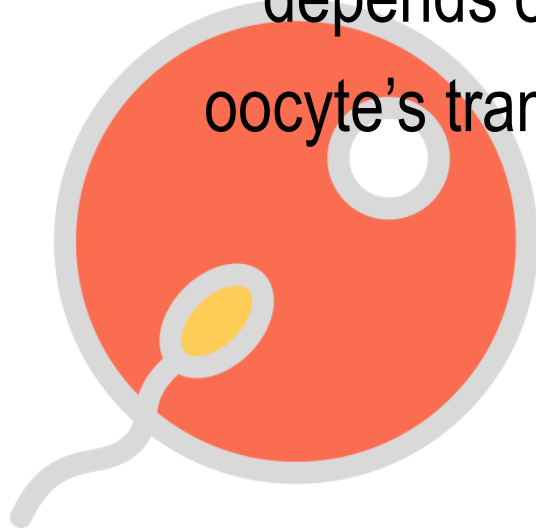


# INTRODUCTION

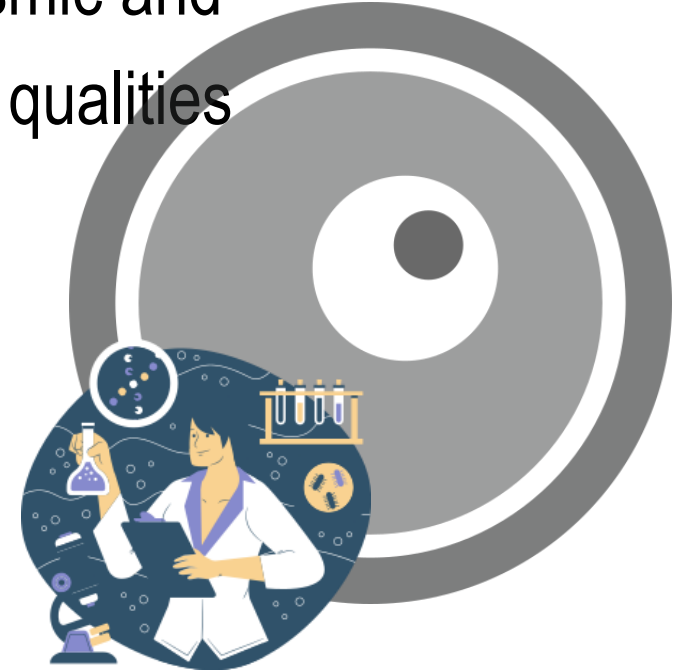


Spermatozoon: do not possess DRA

Once fertilization takes place, DRA depends on the oocyte's transcripts



Cytoplasmic and genomic qualities



Extent of SDF



# INTRODUCTION



Oocyte quality



Oocyte age



## Effect of sperm DNA fragmentation on pregnancy outcome depends on oocyte quality

Marcos Meseguer, Ph.D.,<sup>a</sup> Rebeca Santiso, Ph.D.,<sup>b,c</sup> Nicolas Garrido, Ph.D.,<sup>a</sup> Sandra García-Herrero, Ph.D.,<sup>a</sup> Jose Remohí, M.D.,<sup>a</sup> and Jose Luis Fernandez, M.D.<sup>b,c</sup>

<sup>a</sup>IVI, Universidad de Valencia, Valencia; <sup>b</sup> Sección de Genética y Unidad de Investigación, Hospital "Teresa Herrera," Complejo Hospitalario Universitario A Coruña, A Coruña; and <sup>c</sup> Centro Oncológico de Galicia, A Coruña, Spain

## OBJECTIVE

To investigate the impact of nuclear SDF on the outcomes of ICSI in women of different age ranges.

# MATERIAL AND METHODS

## • *Patients and Experimental design*

Historical cohort study  
June/2017 - Dec/2019

ICSI cycles (n=540)

Maternal age

≤36 years old  
(n=285)

37-40 years old  
(n=147)

> 40 anos  
(n = 108).

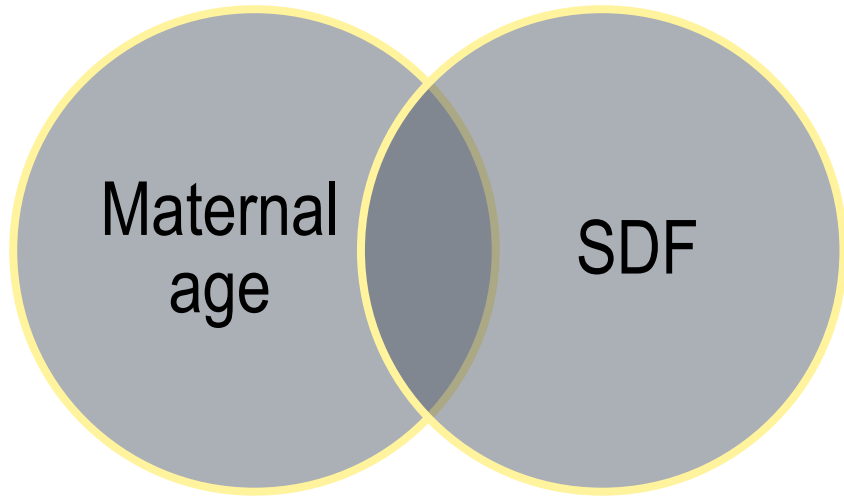
SDF  
(chromatin dispersion test)

low fragmentation index  
(≤30% SDF)

high fragmentation index  
(>30% SDF)

# MATERIAL AND METHODS

- ***Patients and Experimental design***



- *Generalized linear models*
- *Bonferroni post hoc test*
- *Adjustment for potential confounders*



# RESULTS

**GZLM results for the association between SDF and male and female age according to maternal age ranges**

Groups	≤36 years old (n=285)			37-40 years old (n=147)			>40 years old (n=108)		
Age	≤30% SDF (n=171)	>30% SDF (n=114)	p	≤30% SDF (n=99)	>30% SDF (n=48)	p	≤30% SDF (n=64)	>30% SDF (n=44)	p
Paternal	35.6 ± 0.2	36.0 ± 0.3	0.720	38.8 ± 0.2	40.7 ± 0.4	0.036	42.1 ± 0.4	43.7 ± 0.5	0.225
Maternal	32.8 ± 0.1	32.7 ± 0.2	0.746	38.5 ± 0.1	38.6 ± 0.1	0.855	42.7 ± 0.1	42.8 ± 0.2	0.879

# RESULTS

## GZLM results for the association between SDF and response to controlled ovarian stimulation according to maternal age ranges

Groups	≤36 years old (n=285)			37-40 years old (n=147)			>40 years old (n=108)		
	≤30% SDF (n=171)	>30% SDF (n=114)	p	≤30% SDF (n=99)	>30% SDF (n=48)	p	≤30% SDF (n=64)	>30% SDF (n=44)	p
<b>Response to COS</b>									
Dose of FSH	2439.7 ± 25.9	2408.1 ± 41.5	0.747	2615.9 ± 26.9	2551.8 ± 41.3	0.515	2586.7 ± 39.6	2715.4 ± 59.2	0.367
Retrieved oocytes (n)	13.5 ± 0.3	11.9 ± 0.5	0.194	8.6 ± 0.3	9.0 ± 0.5	0.761	5.8 ± 0.4	7.5 ± 0.6	0.201
Mature oocytes (n)	10.3 ± 0.2	8.5 ± 0.4	0.064	6.2 ± 0.5	6.2 ± 0.8	0.973	4.1 ± 0.3	5.5 ± 0.4	0.200

# RESULTS

## GZLM results for the association between SDF and laboratory outcomes according to maternal age ranges

Groups	≤36 years old (n=285)			37-40 years old (n=147)			>40 years old (n=108)		
	≤30% SDF (n=171)	>30% SDF (n=114)	p	≤30% SDF (n=99)	>30% SDF (n=48)	p	≤30% SDF (n=64)	>30% SDF (n=44)	p
<i>Lab results</i>									
Fertilization	89.1	89.5	0.645	79.4	83.4	0.356	78.3	76.5	0.745
High-quality embryos	42.6	42.5	0.977	38.1	36.0	0.676	54.4	33.1	<b>0.005</b>
Blastocyst development	85.3	84.2	0.645	42.6	51.6	0.224	49.6	30.2	<b>0.035</b>
Embryos transferred	1.2 ± 0.2	1.2 ± 0.3	0.843	1.1 ± 0.1	1.1 ± 0.2	0.473	1.1 ± 0.1	1.2 ± 0.1	0.789

# RESULTS

## GZLM results for the association between SDF and clinical outcomes according to maternal age ranges

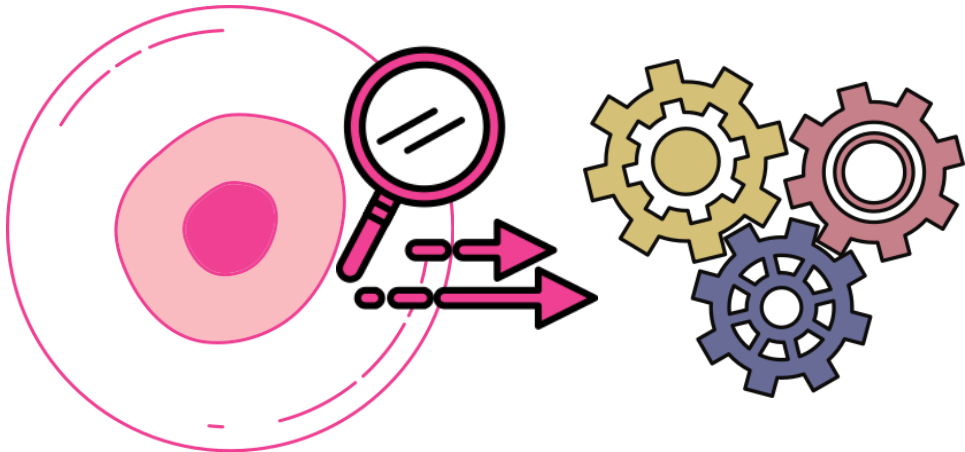
Groups	≤36 years old (n=285)			37-40 years old (n=147)			>40 years old (n=108)		
<i>Clinical results</i>	≤30% SDF (n=171)	>30% SDF (n=114)	p	≤30% SDF (n=99)	>30% SDF (n=48)	p	≤30% SDF (n=64)	>30% SDF (n=44)	p
<b>Implantation rate (%)</b>	42.3	41.5	0.880	28.9	30.6	0.757	19.7	11.9	<b>&lt;0.001</b>
<b>Pregnancy rate (%)</b>	40.0	39.1	0.840	27.7	28.6	0.781	20.0	7.7	<b>0.040</b>
<b>Miscarriage rate (%)</b>	9.3	11.1	0.665	31.2	22.2	0.875	12.5	100	<b>&lt;0.001</b>

# DISCUSSION



AGE

Ageing compromises the oocyte's DNA repair activity

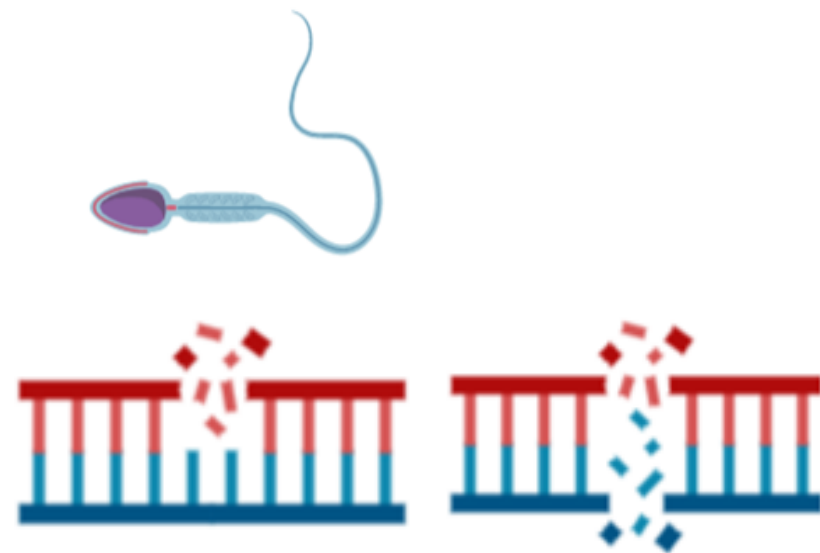


Oocytes from the study were in fact provided with DNA repair activity or to what extent the DNA of injected spermatozoa was damaged ?

# DISCUSSION



AGE



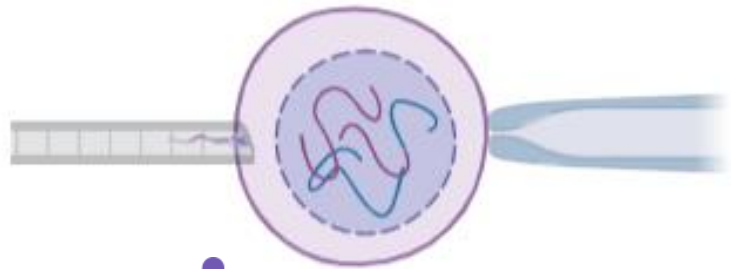
> 30% de SDF

**X**

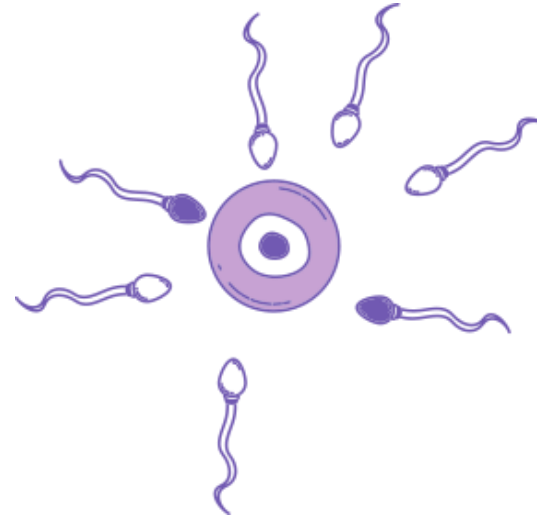
> 40 y-old



# DISCUSSION



VS

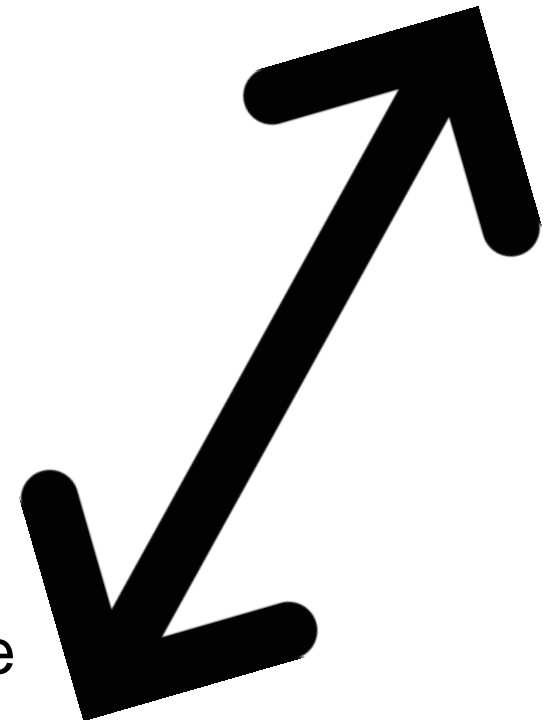


The higher the chance  
of selecting one with  
fragmented DNA

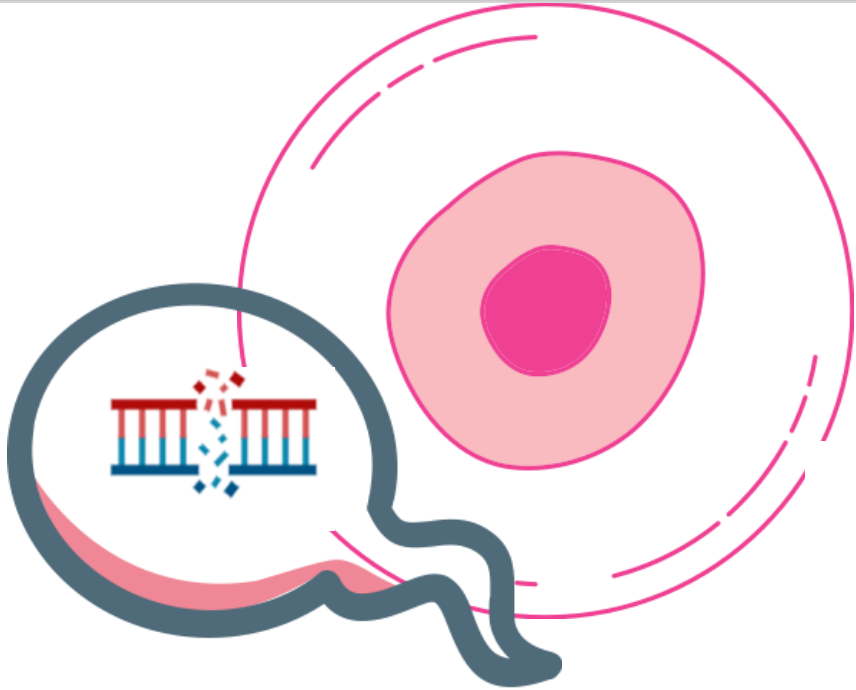


Sperm with hidden  
abnormalities may be  
selected for injection

The higher the  
SDF index



# DISCUSSION



Apoptosis, which will destroy one or more blastomeres, reducing embryo viability



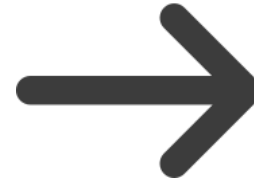
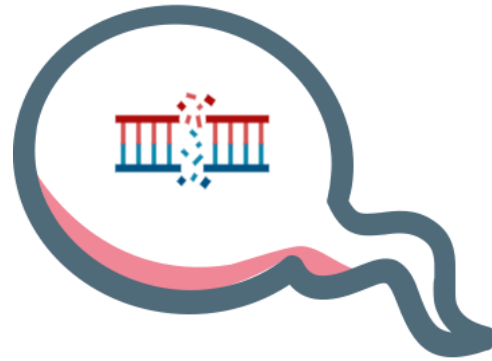
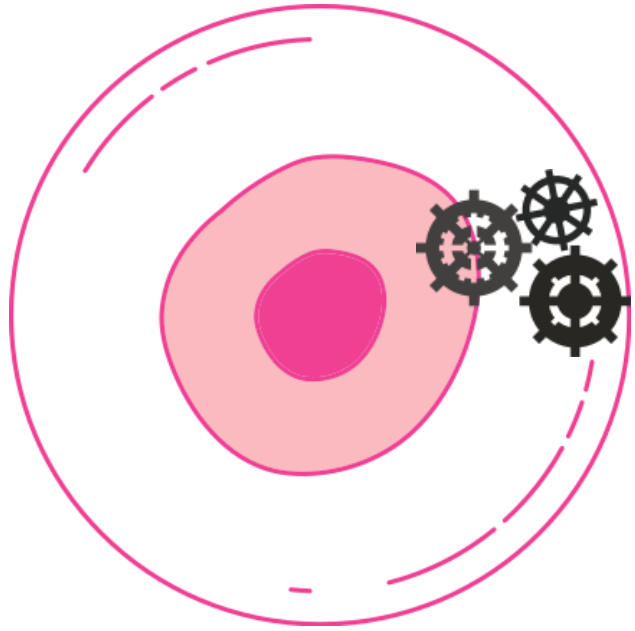
Tolerate the lesion, which may lead to mutation and eventual carcinogenesis in the offspring



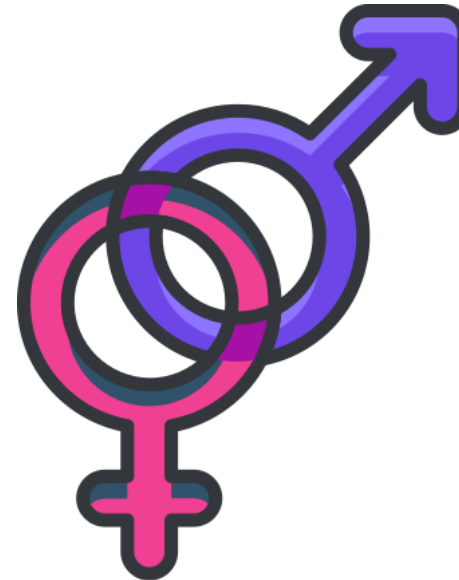
Repair the lesion



# DISCUSSION



Once the oocyte is provided with repair activity, the impact of SDF on reproductive outcomes is a combination of both male and female factors



# DISCUSSION



Ability to cope  
with SDF

Advanced maternal age is  
associated with reduced  
oocytes' mRNA stores and  
DRA

*Human Molecular Genetics*, 2004, Vol. 13, No. 19 2263–2278  
doi:10.1093/hmg/ddh241  
Advance Access published on August 18, 2004

## Age-associated alteration of gene expression patterns in mouse oocytes

Toshio Hamatani<sup>†</sup>, Geppino Falco<sup>†</sup>, Mark G. Carter, Hidenori Akutsu, Carole A. Stagg,  
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Developmental Genomics and Aging Section, Laboratory of Genetics, National Institute on Aging,  
National Institutes of Health, 333 Cassell Drive, Suite 3000, Baltimore, MD 21224, USA

Received May 9, 2004; Revised and Accepted July 20, 2004

## CONCLUSIONS

- ✓ High SDF index leads to lower implantation and pregnancy rates, and higher miscarriage rate, in ICSI cycles of women with advanced maternal age
- ✓ The same is not observed when maternal age is < 40 years old
- ✓ Ageing may affect the oocyte DRA, leading to the development of an embryo with poor development and implantation potential, when the oocyte is injected with a DNA-damaged spermatozoon

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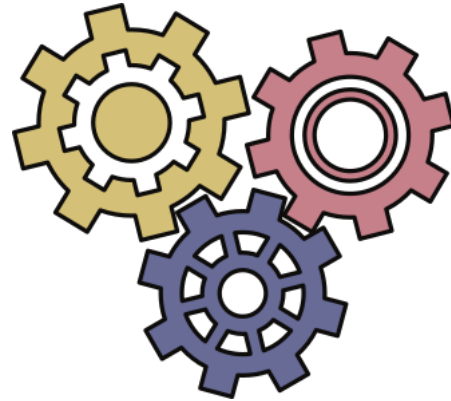
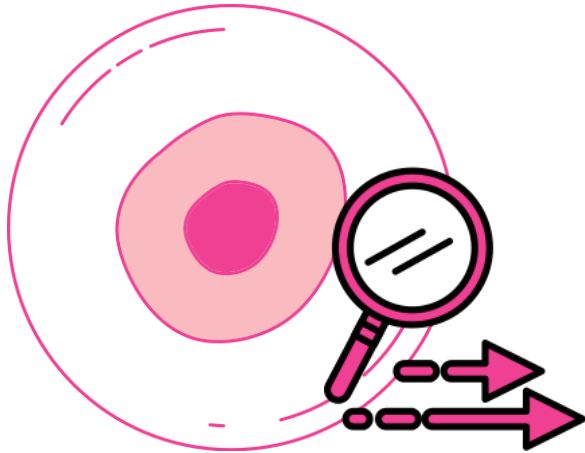
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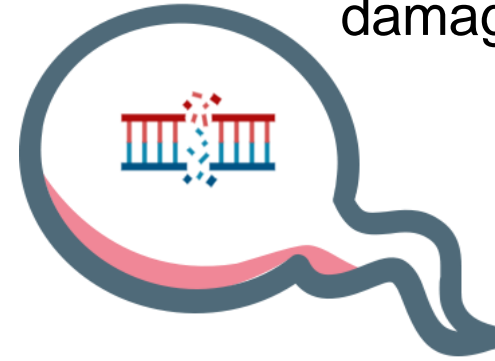
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Janaína Gomes Pinho da Silva  
Katia Rodrigues  
Lucácio de Souza Anjos

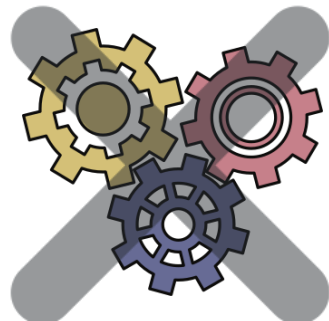
# DISCUSSION



Oocytes from the study were in fact provided with DNA repair activity or to what extent the DNA of injected spermatozoa was damaged ?






AGE



oocytes from women with advanced maternal age were less likely to repair SDF, thus resulting in lower pregnancy outcomes.

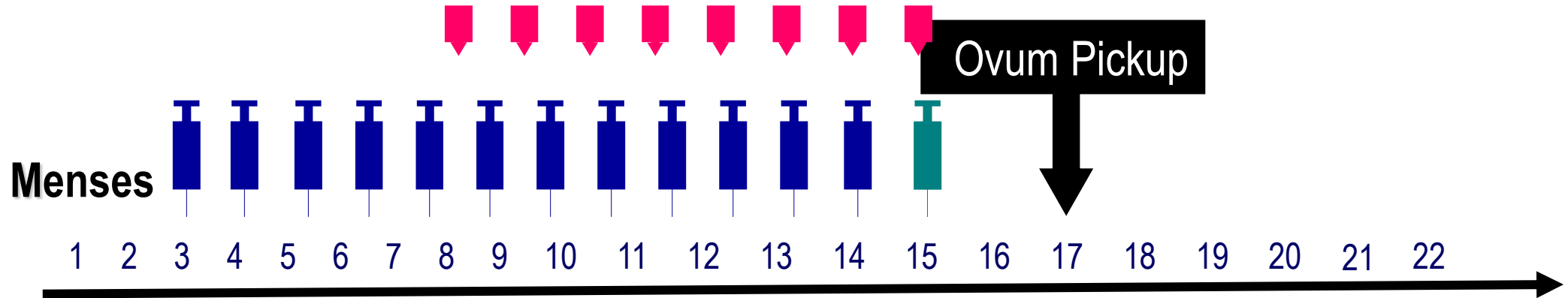
# MATERIAL AND METHODS

- Controlled ovarian stimulation***

-  GnRH Antagonist
-  Recombinant FSH
-  Recombinant hCG



E2



# INTRODUCTION



Asian Journal of Andrology (2017) 19, 80–90  
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www.asiaandro.com; www.ajandrology.com



Open Access

ORIGINAL ARTICLE

**A systematic review and meta-analysis to determine the effect of sperm DNA damage on *in vitro* fertilization and intracytoplasmic sperm injection outcome**

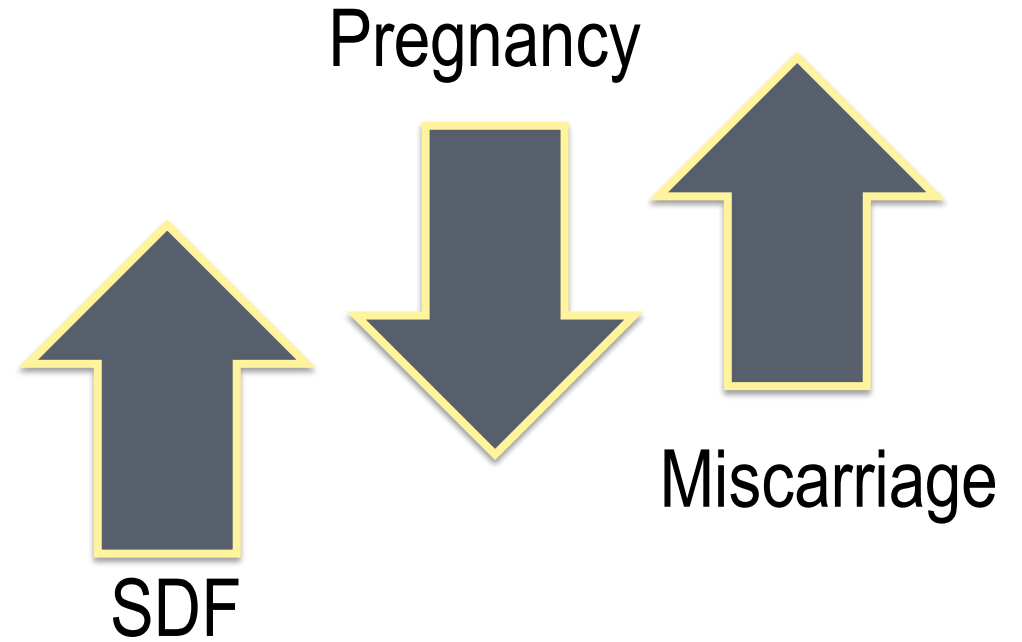
Luke Simon<sup>1\*</sup>, Armand Zini<sup>2\*</sup>, Alina Dyachenko<sup>2</sup>, Antonio Ciampi<sup>2</sup>, Douglas T Carrell<sup>1,3,4</sup>



## Sperm DNA fragmentation and recurrent pregnancy loss: a systematic review and meta-analysis

Dana B. McQueen, M.D., M.A.S., John Zhang, Ph.D., and Jared C. Robins, M.D.

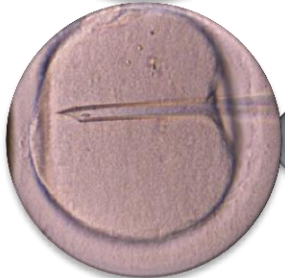
Division of Reproductive Endocrinology and Infertility, Department of Obstetrics and Gynecology, Northwestern University, Chicago, Illinois



# MATERIAL AND METHODS



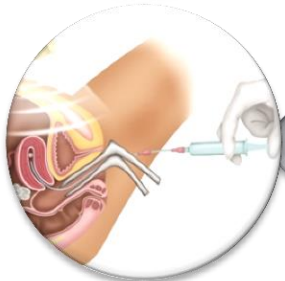
Incubation, denudation and nuclear maturation evaluation



ICSI - (Palermo et al., 1992)



Embryo culture until day 5



One or two blastocysts transferred



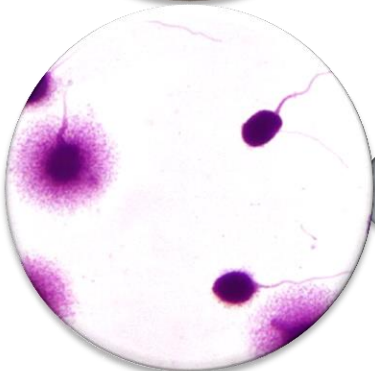
# MATERIAL AND METHODS



Semen samples were evaluated according to the threshold values established by the WHO in 2010



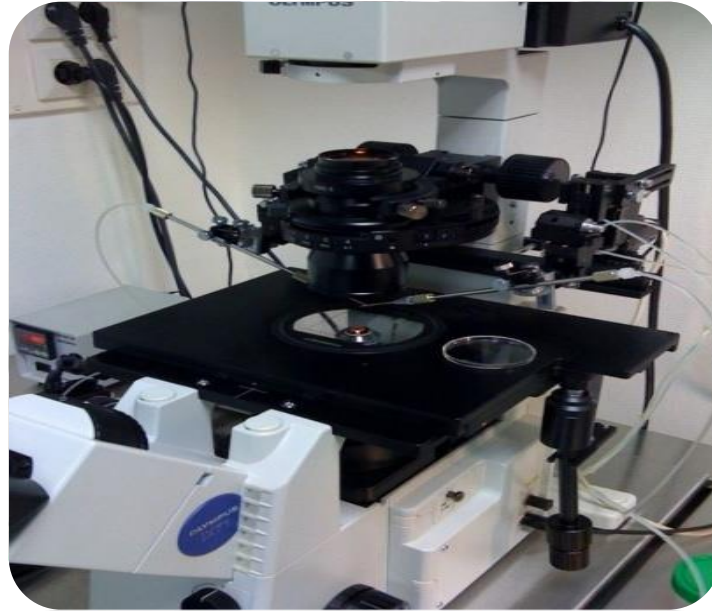
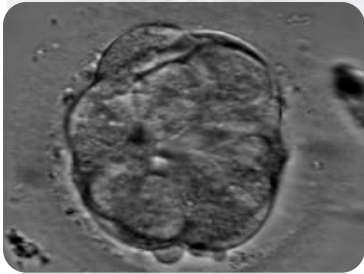
Sperm preparation: 2-layered density gradient centrifugation technique



The sperm DNA fragmentation: Sperm chromatin dispersion test

# MATERIAL AND METHODS

- *Embryo morphology and transfer*



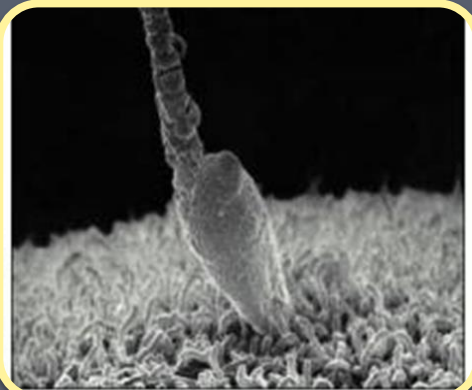
# INTRODUCTION



Studies showing a correlation between semen quality and reproductive outcomes



So far, there is no predictive threshold for success for conventional semen parameters



## Spermatozoa

- subcellular or nuclear factors that are not recognized during conventional semen analysis,
- May contribute to male factor infertility