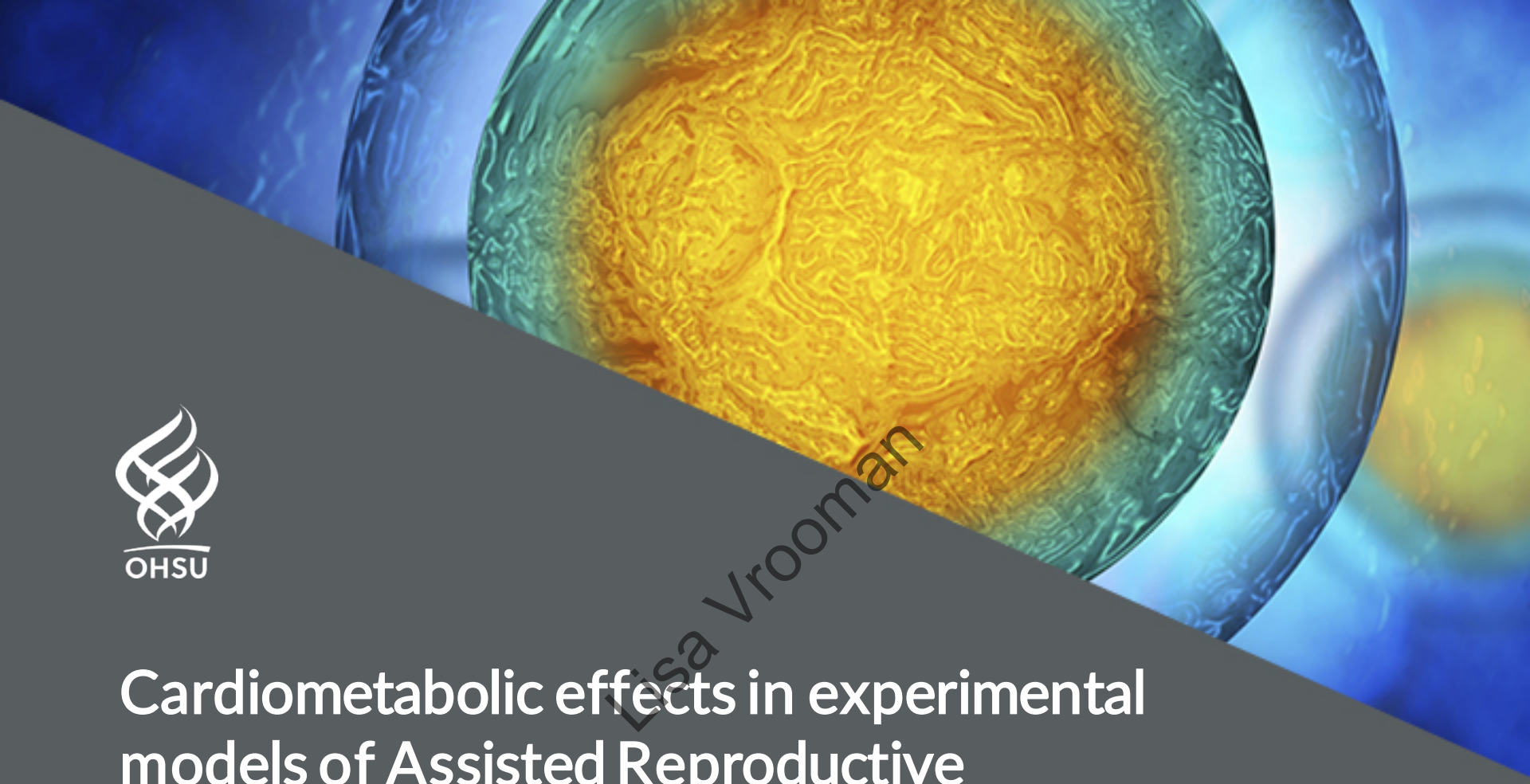
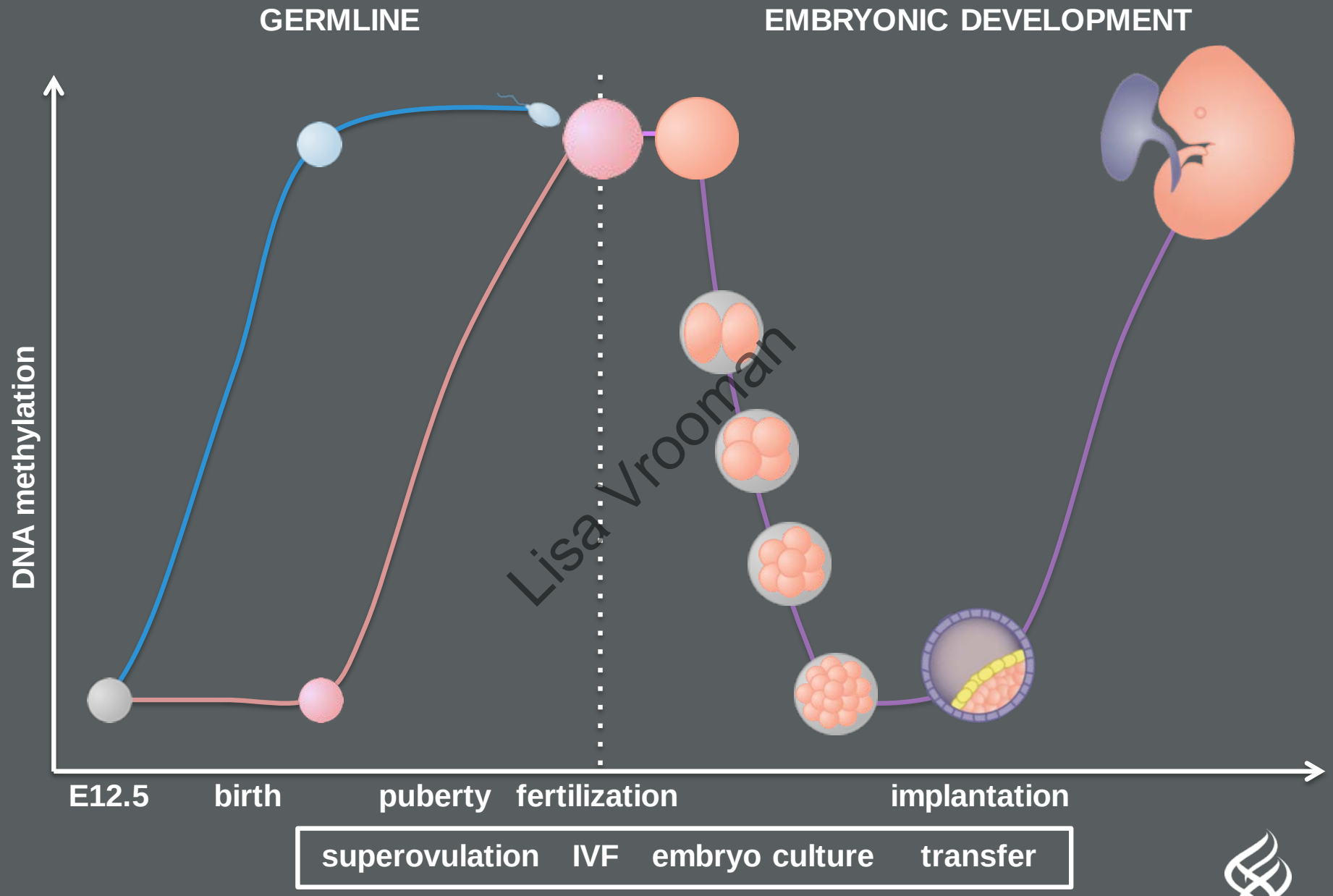


Cardiometabolic effects in experimental models of Assisted Reproductive Technologies

Lisa Vrooman, PhD | August 5, 2021

Developmental origin of health and disease (DOHaD)

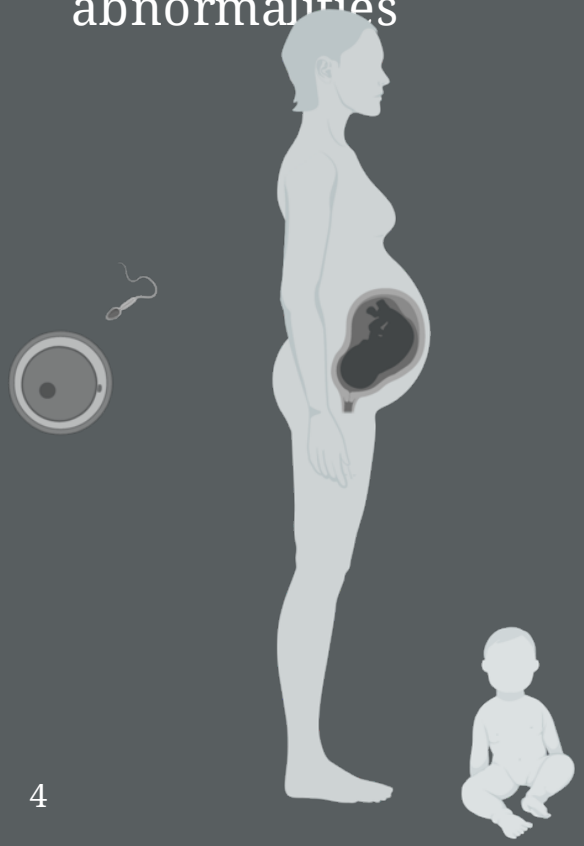
- Research demonstrating the link between maternal, perinatal, and early childhood factors and risk on non-communicable diseases
- Are ARTs a critical window of 'exposure'?


GERMLINE
EMBRYONIC DEVELOPMENT
DNA methylation
Lisa Vrooman
E12.5
birth
puberty
fertilization
implantation
superovulation
IVF
embryo culture
transfer

Developmental origin of health and disease (DOHaD)

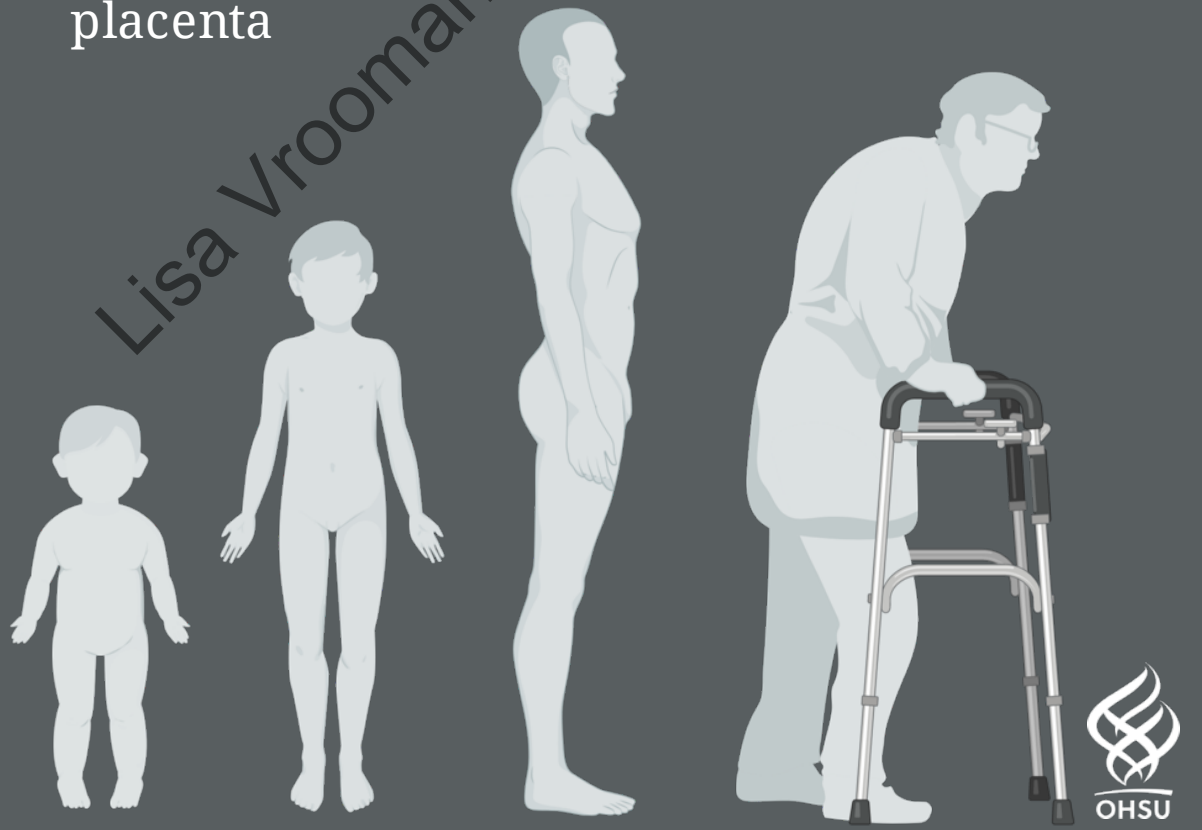
SEVERE

- Infertility
- Pregnancy loss
- Congenital abnormalities



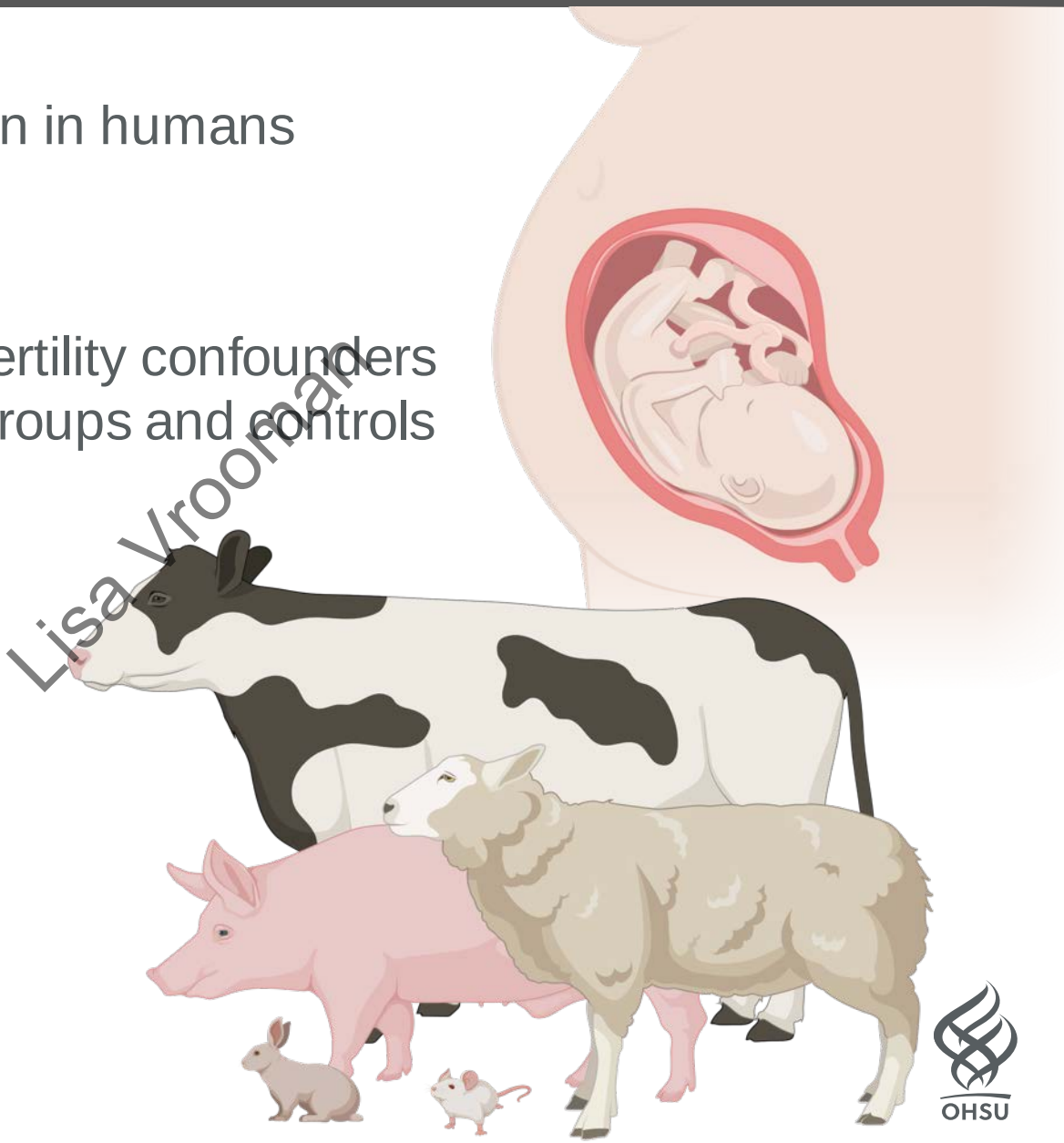
MILD

- Relatively normal postnatal development
- Risk for chronic diseases later in life
- Directly affect fetus or indirectly affect fetus via placenta



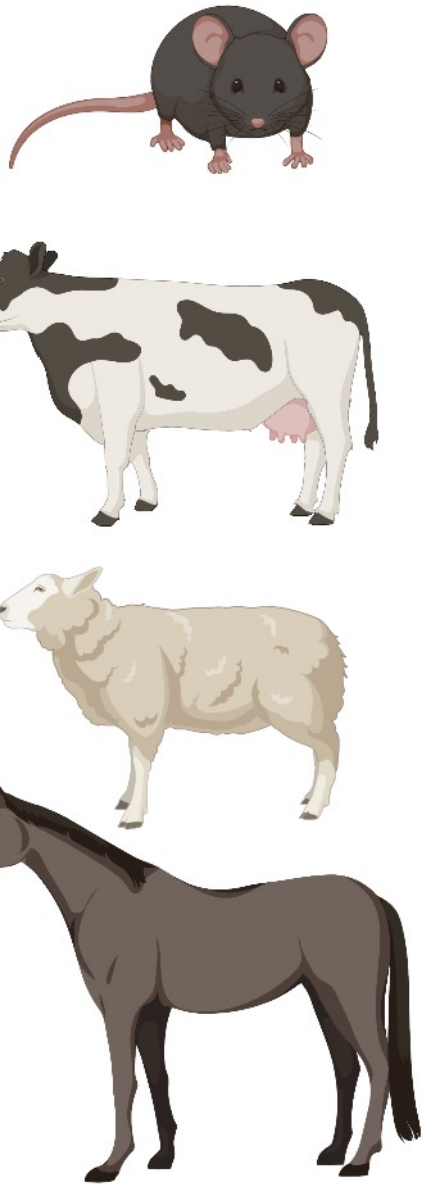
Benefit of Animal Models

- Ethics of experimentation in humans
- Controlled environment
- Controlled genetics
- Removes underlying infertility confounders between experimental groups and controls



Most prevalent animal models of ART

- Mouse
- Cow
- Sheep
- Horse



Most prevalent animal models of ART

- Mouse
- Cow
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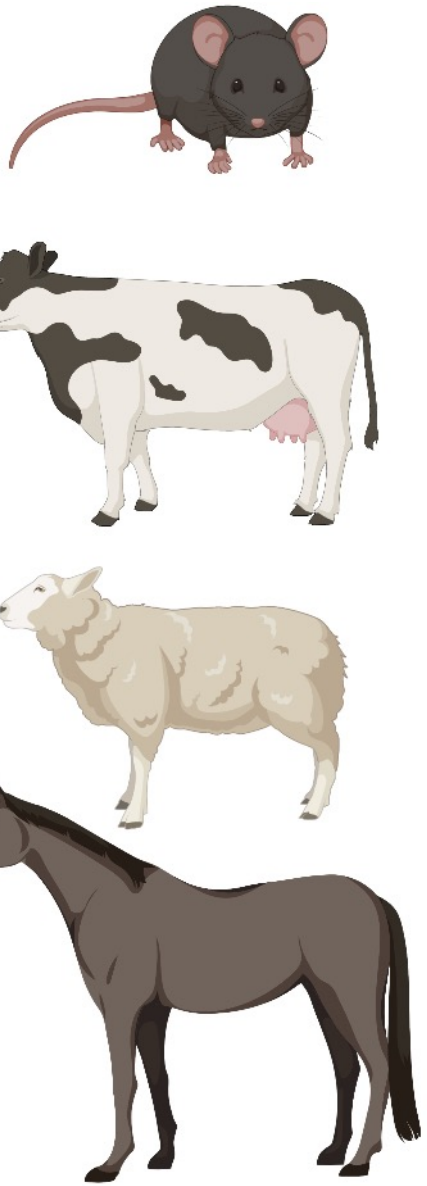
- Due to complex nature of studying long-term effects, in vitro and computer models are inadequate



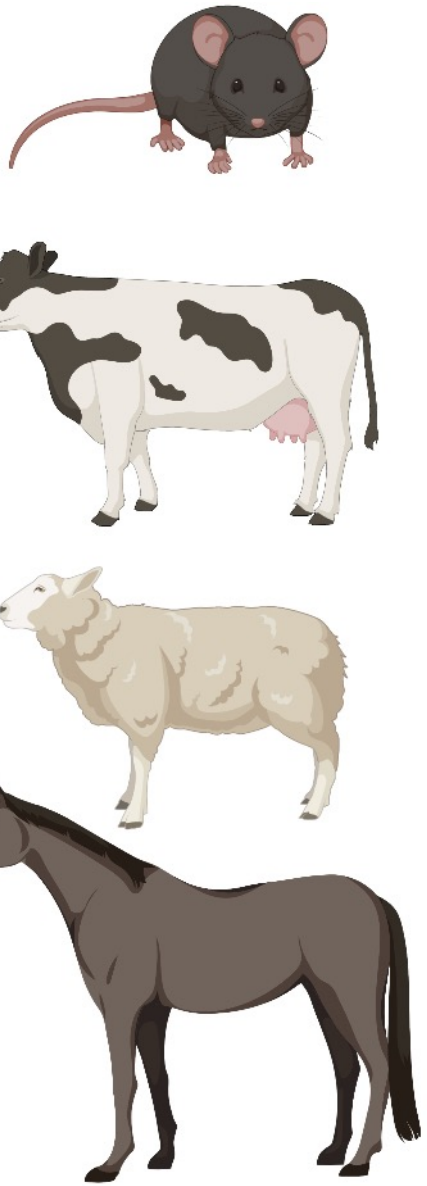
Most prevalent animal models of ART

- Mouse
- Cow
- Sheep
- Horse

- Due to complex nature of studying long-term effects, in vitro and computer models are inadequate

Long-term health outcomes

- **MOUSE**



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Critical reading of animal studies

- Hormone levels
- Suboptimal culture methods
  - Oxygen levels
  - Type of culture media
- Sexes analyzed
  - Only one sex?
  - Both sexes together?
- Age at analysis
- Litter size

Prevention of vascular dysfunction and arterial hypertension in mice generated by assisted reproductive technologies by addition of melatonin to culture media

Am J Physiol Heart Circ Physiol 309: H1151–H1156, 2015.
First published August 14, 2015; doi:10.1152/ajpheart.00621.2014.

Emrush Rexhaj,[1] Agim Pireva,[1] Ariane ...
Pierre Dessen,[3] Claudia ...

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DOI 10.1095/biolreprod.113.113134

In Vitro Fertilization Affects Growth and Glucose Metabolism in a Sex-Specific Manner in an Outbred Mouse Model[1]

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RESEARCH ARTICLE

FASEB JOURNAL

Sex-specific effects of in vitro fertilization on adult metabolic outcomes and hepatic transcriptome and proteome in mouse

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Abstract
Although in vitro fertilization (IVF) is associated with adverse perinatal outcomes, there is increasing concern about the long-term and sex-specific health implications. Augmenting our IVF mouse model to longitudinally investigate metabolic outcomes in offspring from optimal neonatal litter sizes, we found sex-specific metabolic outcomes in IVF offspring. IVF-conceived females had higher body weight and cholesterol levels compared to naturally conceived females, whereas IVF-conceived males had higher levels of triglycerides and insulin, and increased body fat composition. Through adult liver transcriptomics and proteomics, we identified sexually dimorphic dysregulation of the sterol regulatory element-binding protein (SREBP) pathways that are associated with the sex-specific phenotypes. We also found that global loss of DNA methylation in placenta was linked to higher cholesterol levels in IVF-conceived females. Our findings indicate that IVF procedures have long-lasting sex-specific effects on metabolic health of offspring and lay the foundation to utilize the placenta as a predictor of long-term outcomes.

KEYWORDS
assisted reproductive technologies, developmental origins of health and disease, long-term health, metabolic outcomes, sex-specific

1 | INTRODUCTION

Over the past 40 years, assisted reproductive technologies (ART), including in vitro fertilization (IVF), have become a highly successful treatment for infertility. Globally, there has been a consistent and steady increase in the use of ART,[1] now contributing to over 9 million births worldwide.[2] Although the majority of IVF births are uncomplicated, IVF is associated with

Abbreviations: ART, assisted reproductive technologies; DEG, differentially expressed gene; DNAm, deoxyribonucleic acid methylation; IPGTT, intraperitoneal glucose tolerance test; IVF, in vitro fertilization; Nat, naturally conceived.

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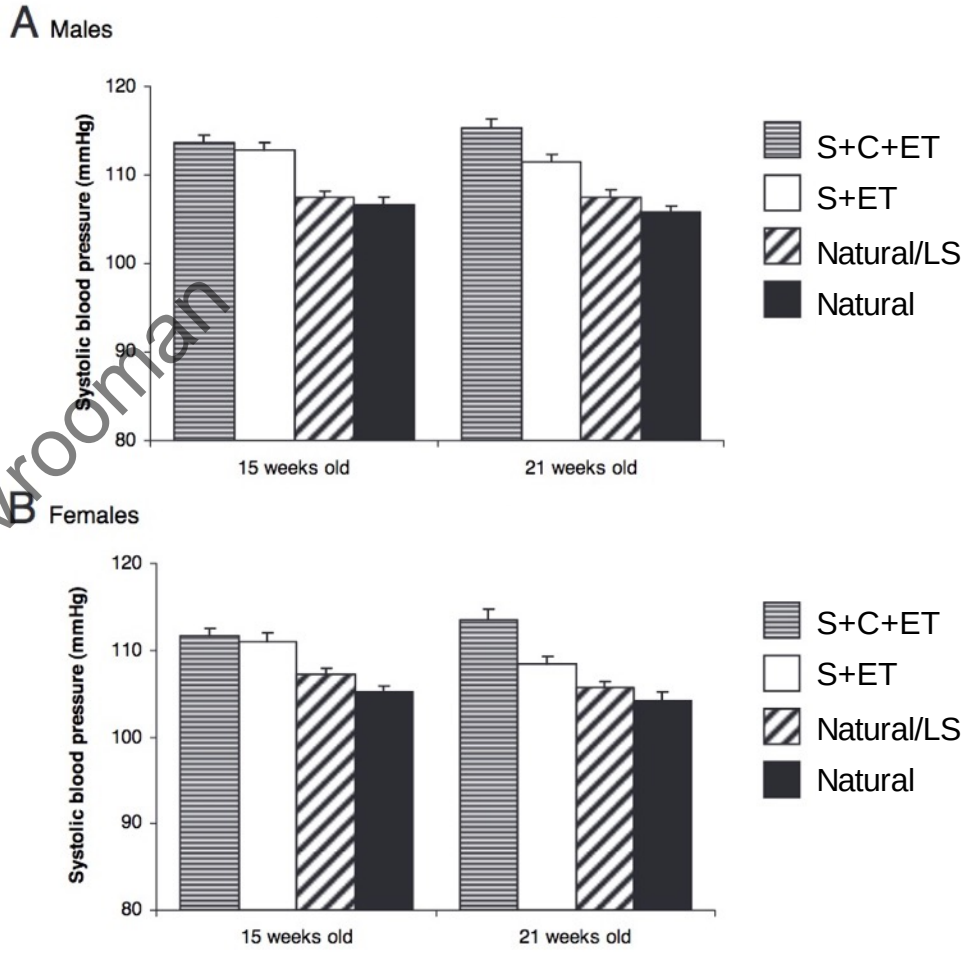
Cardiac outcomes

Watkins et al. 2007

(both sexes)

- Increased blood pressure in males and females that were cultured as preimplantation embryos (fertilization in vivo)



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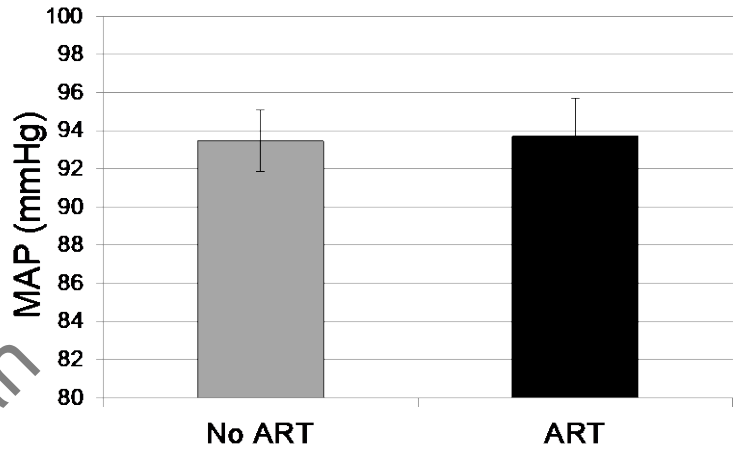
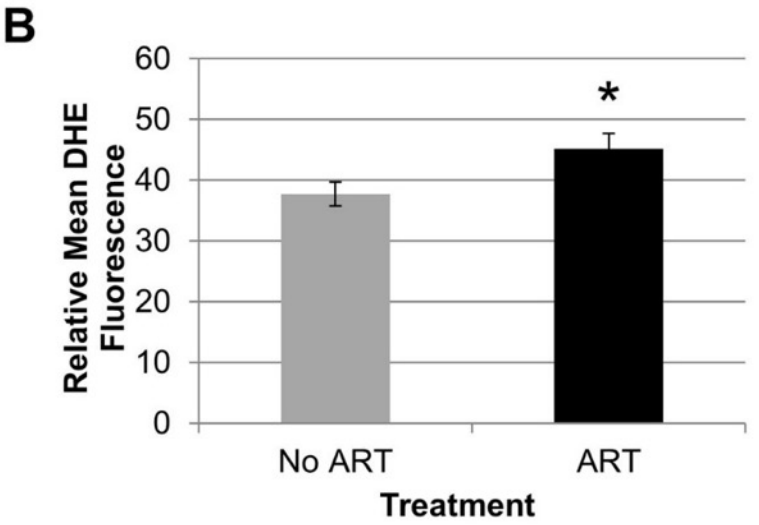
Cardiac outcomes

Schenewerk et al. 2013

(both sexes)

- In juvenile mice produced after suboptimal media culture (fertilization in vivo) with normal or high fat diet:
- No differences in blood pressure
- Increased oxidative stress in mesenteric resistance arteries after culture regardless of diet
- Increased oxidative stress is linked to vascular remodeling which could lead to vascular dysfunction





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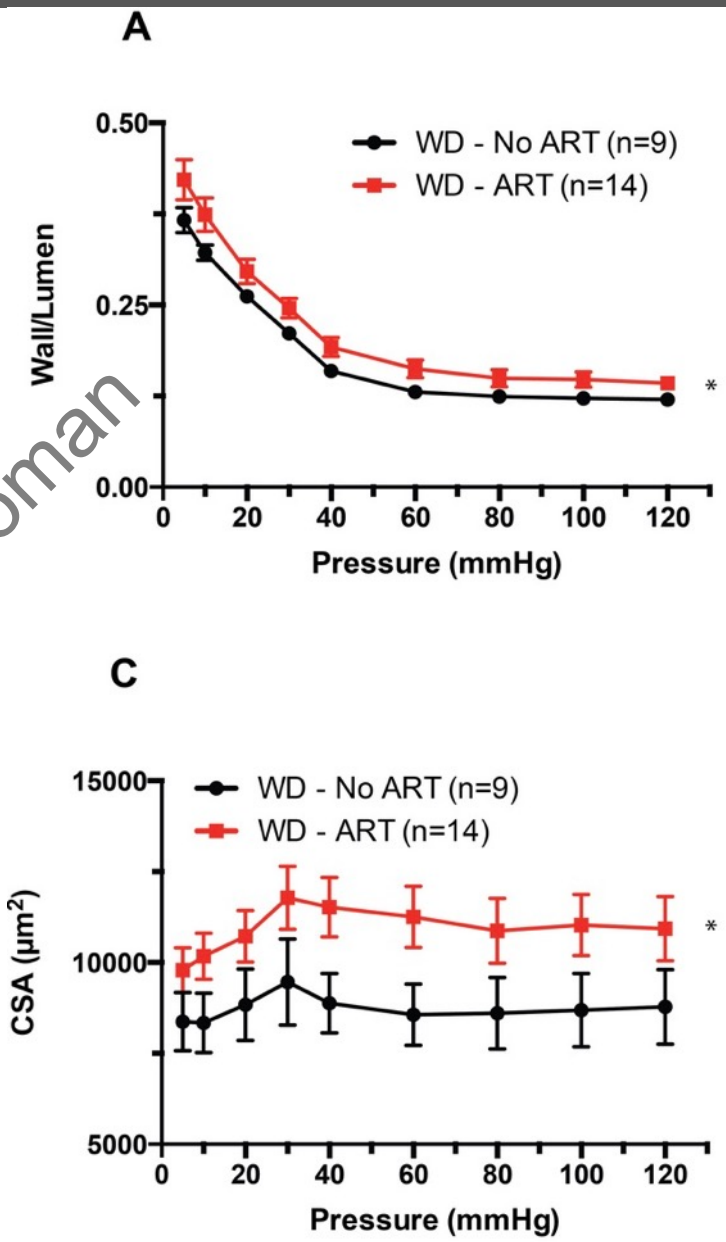
Cardiac outcomes

Ramirez-Perez et al. 2014

(both sexes, analyzed together)

In juvenile mice produced after suboptimal media culture (fertilization in vivo) with normal or high fat diet:

- Mesenteric artery dysfunction and vascular remodeling in cultured, high fat diet mice.



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Cardiac outcomes

Donjacour et al. 2014

(both sexes)

- Reduced blood pressure and enlarged left hearts in males with suboptimal culture as preimplantation embryos

TABLE 3. Cardiovascular phenotype.

Parameter	FB[a]	IVF_{WM}[b]
Heart weight (mg)	144 ± 40	161 ± 31
Systolic blood pressure (mm Hg)	146 ± 12	135 ± 9*
Diastolic blood pressure (mm Hg)	117 ± 13	107 ± 15
Mean blood pressure (mm Hg)	127 ± 15	117 ± 13
Systolic volume (SV, µl)	47 ± 8	55 ± 10
End-diastolic volume (EDV, µl)	57 ± 10	70 ± 14**
End-systolic volume (ESV, µl)	10 ± 1	14 ± 4**
Ejection fraction (EF, %)	85 ± 1	84 ± 3
Cardiac output (CO, ml/min)	33 ± 6	35 ± 6
Fraction shortening (FS)	48 ± 2	47 ± 3
Ventricular mass (g)	142 ± 24	181 ± 39**
Left-ventricular intraseptal wall dimension at diastole (mm)	3.7 ± 0.3	4.0 ± 0.4**
Left-ventricular posterior wall dimension at diastole (mm)	0.7 ± 0.1	0.8 ± 0.1**

[a] n = number ranged from 11–13, from three litters.
[b] n = number ranged from 9–10, from four litters.
* Significantly lower than FB.
** Significantly greater than FB.

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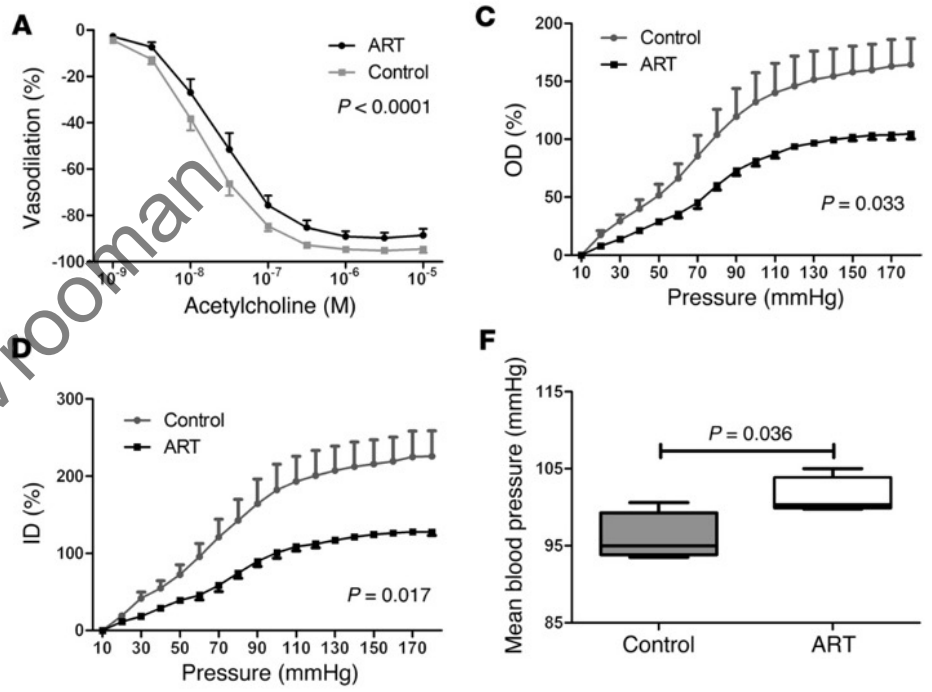
Cardiac outcomes

Rexhaj et al. 2015

(males only)

- Impaired endothelial-dependent artery vasodilation
- Increased cartotid artery stiffness
- Increased blood pressure
- Administration of butyrate (deacetylase inhibitor) improved vascular function, suggesting dysfunction is linked to epigenetic changes.



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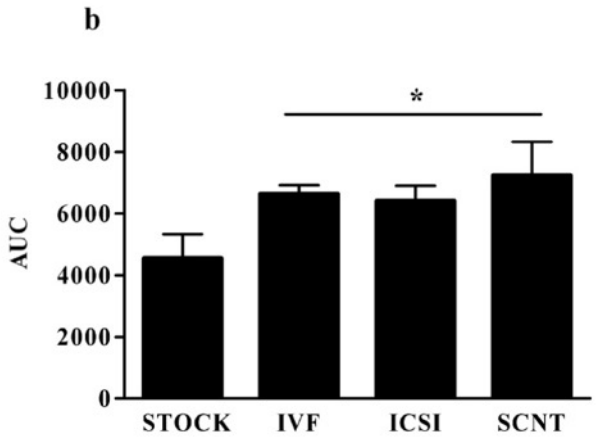
Metabolic outcomes

- Conflicting findings and sex-dependent differences

Scott et al. 2010

(both sexes)

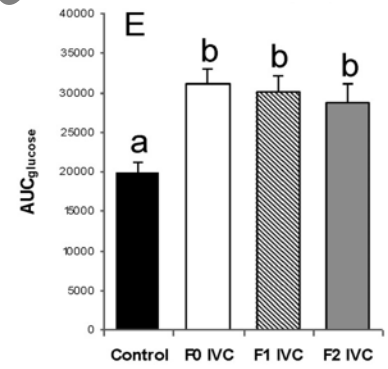
- Impaired glucose tolerance in females conceived by ICSI or IVF, no changes in males

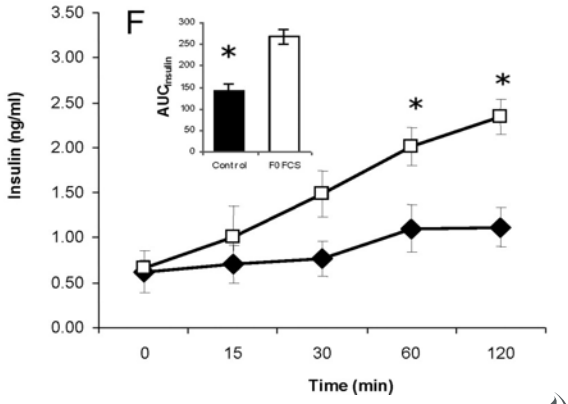


Calle et al. 2012

(males only)

- Impaired glucose homeostasis in males after embryo culture
- Transmitted to offspring





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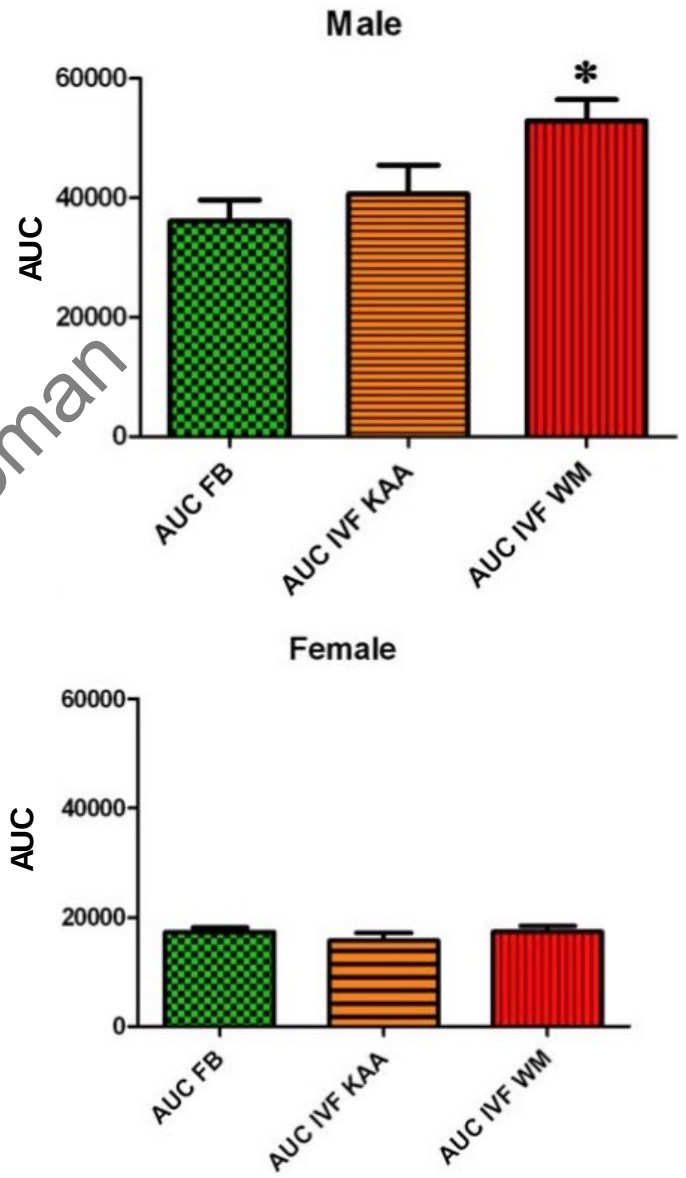
Metabolic outcomes

Donjacour et al. 2014

(both sexes)

- Impaired glucose homeostasis in males after suboptimal embryo culture, no changes in females. Both sexes normal under optimal conditions.



Metabolic outcomes

Feuer et al. 2014

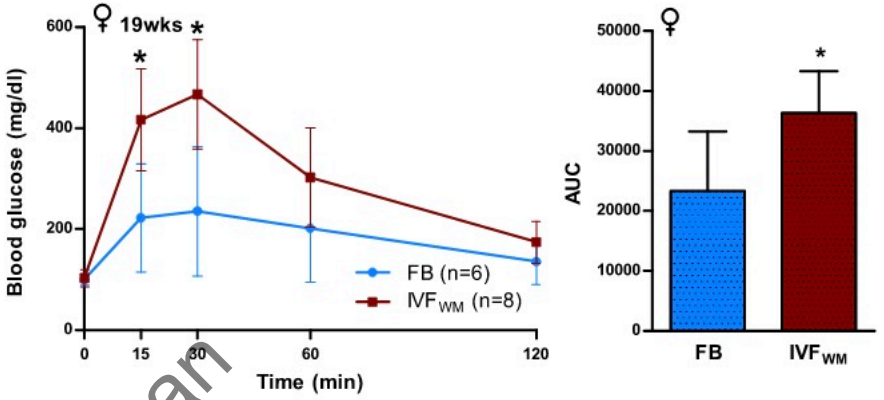
(both sexes)

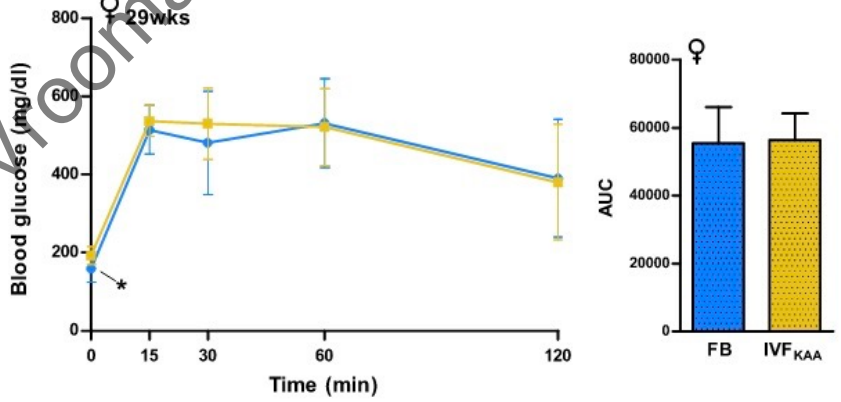
IVF under suboptimal condition

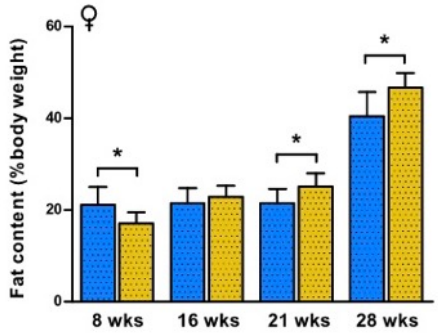
- Impaired glucose tolerance in females; male trend but not significant
- Reduced BMI in males and females

IVF under optimal conditions:

- Increased fat deposition and fasting glucose levels in females, but glucose tolerance was normal
- No differences in males







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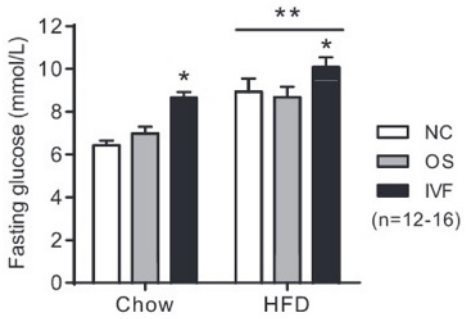
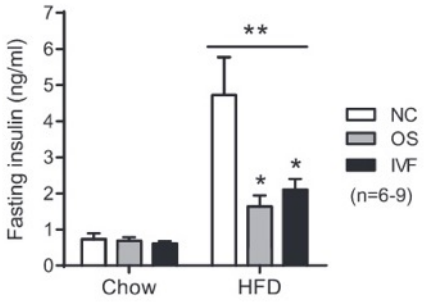
Metabolic outcomes

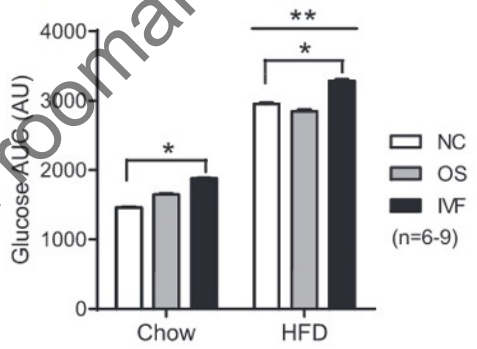
Chen et al. 2014a and 2014b

Males

- Increased fasting glucose levels and impaired glucose tolerance in IVF males with normal or high fat diet
- Reduced insulin levels in IVF males on high fat diet only

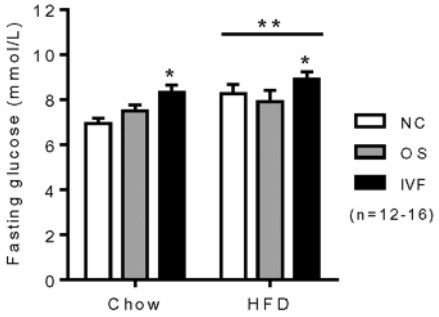


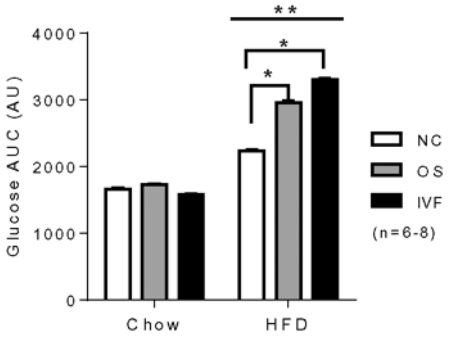




Females

- Increased fasting glucose levels and impaired glucose tolerance in IVF and hormone stimulated groups with high fat diet.





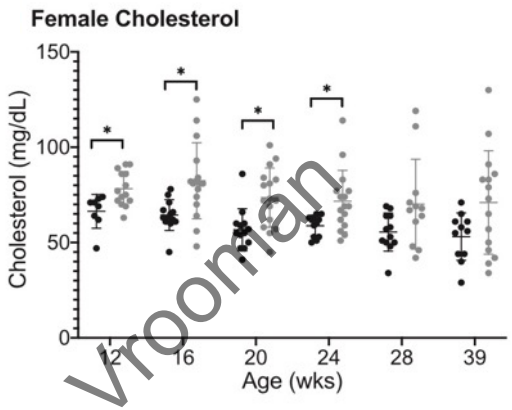
Metabolic outcomes

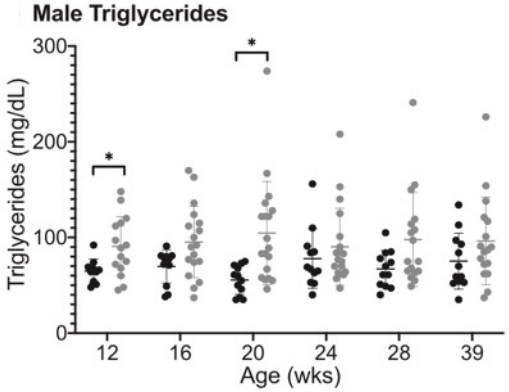
Narapareddy et al. 2021

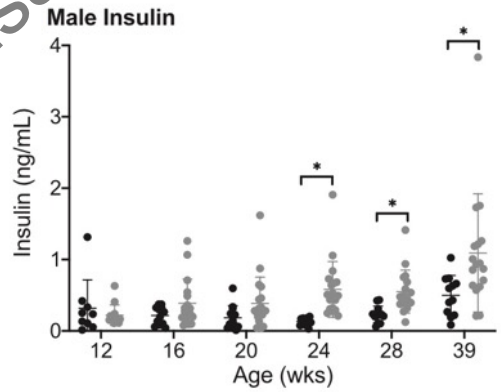
(both sexes)

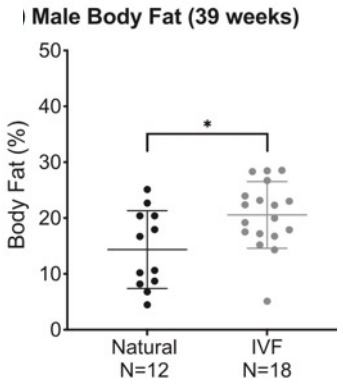
IVF under optimal conditions:

- Increased body weight and cholesterol in females
- Increased triglycerides, insulin, and body fat in males
- No changes in blood pressure









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Other outcomes

Lifespan

Sommovilla et al. 2005 (both sexes)

- No differences

Rexhaj et al. 2013 (males only)

- IVF mice had 25% reduced lifespan with high fat diet

Neurological outcomes

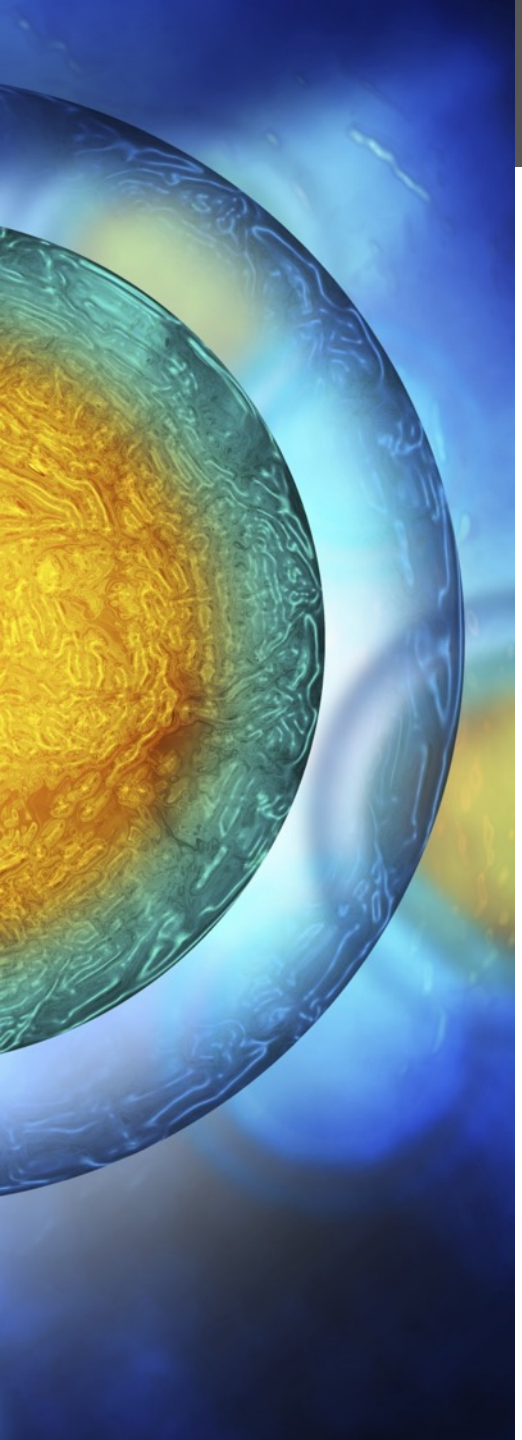
Ecker et al. 2004 (males only) and Fernandez-Gonzalez et al. 2004

- Decreased anxiety and impaired spatial memory in mice after suboptimal embryo culture

Other outcomes

Fernandez-Gonzalez. 2004 (males only)

- Increased organ size
- Risk of developing pneumonia, steatosis, and kidney inflammation



The Future

Add-on procedures

- Cryopreservation
- Trophectoderm biopsy

Techniques

- ‘omics’ analyses to determine mechanisms
  - Transcriptome
  - Proteome
  - Epigenome
  - Metabolome

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Thank You