Basic science session:

Genome instability in human oocytes and embryos

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- Data protection license (GDPR): SUND-2016-50
- Declare no competing interests
Genetic diversity and genome instability

- > 7,000 genetic disorders
- 50-75% with impact on pediatric health
- Majority are rare (< 1:2,000)
- 6% of children have a genetic predisposition to ‘serious’ disease

(Wright et al., Nat Gen 2017)
>7,000 genomic disorders

Technology-driven discovery

variants (SNPs)

gene conversions
de novo mutations
insertions/deletions (CNV)
structural rearrangements
recombination
aneuploidies

NGS

SNP arrays

(Blanshard et al., Methods in Mol. Biol., 2018)
Germline: origins of genome instability

- **Meiosis (& PGC)**: sperm → mature MII → zygote
- **Embryonic mitosis**: cleavage stage → blastocyst
- **Tissue differentiation**: “black box”
- **Birth**: fetus → births
Hypothesis: understand the basics and translate

“Valley of Death”

(Butler, Nature, 2008)
Events in pregnancy losses and live births

Major of aneuploidy in pregnancy losses and live births are whole chromosomes from meiosis

Natural fertility in human is ‘inefficient’

Whole chromosome aneuploidy shapes natural fertility

Meiotic errors in oocytes follow a U shape

Error types differ w. age

Chromosomes afflicted differ

Cohesion loss drives aneuploidy in AMAs

Very young females?

Idea is that meiotic errors result in uniform, aneuploid embryo.

Euploid or uniform **aneuploid**

Selection (PGT-A: noninvasive)

Improve clinical endpoints  
(live births, PL, reduce twin pregnancies)
Heritability of in- and subfertility?

Common phenotype: common variants, each with small effect

Rare, pathogenic mutations, with high penetrance (explain very few cases- e.g. TUBB8)
Genetic regulation of aneuploidy in human eggs?

“A candidate gene analysis and GWAS for genes associated with maternal nondisjunction of chromosome 21”

Jonathan M. Chernus, Emily G. Allen, Zhen Zeng, Eva R. Hoffman, Terry J. Hassold, Eleanor Feingold, Stephanie L. Sherman

(PLoS Genet., in press)
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“A candidate gene analysis and GWAS for genes associated with maternal nondisjunction of chromosome 21”

Jonathan M. Chernus¹, Emily G. Allen², Zhen Zeng³, Eva R. Hoffman⁴, Terry J. Hassold⁵, Eleanor Feingold¹,³*,¶, Stephanie L. Sherman²

Translational perspective: Polygenic risk scores?

(PLoS Genet., in press)
Current model for maternal meiotic errors in AMA

(Capablo, Hoffmann et al, HRU, 2017)
Ottolini et al., Nat Genet, 2015)
Recombination counteracts cohesion loss (with age)

(Matplotlib diagram showing the process of meiosis and development from meiosis to embryo)

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Recombination counteracts cohesion loss (with age)

> 200 genes

(Capablo, Hoffmann *et al.*, HRU, 2017)
(Ottolini *et al.*, *Nat Genet*, 2015)

Maternal age
‘cohesion weakening/loss’, ‘epigenetic drift’

> 80 genes

(Capablo, Hoffmann *et al.*, HRU, 2017)
(Ottolini *et al.*, *Nat Genet*, 2015)
Clinical utility?

- Personalized risk?
- Aging features?
  (surrogate, biomarker)
- Genetics?

Mosaicism

**Euploid or uniform aneuploid**

**Mosaic (proportion of affected embryo)**
Mosaicism and segmental variations

Segmental aneuploidy: part of a chromosome

<table>
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<tr>
<th>whole</th>
<th>&quot;segmental&quot; aneuploidies</th>
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<td>(gross chromosomal rearrangements -&gt; INDELS)</td>
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- arm
- INDEL
- translocation
- chromothripsis

- resolution
- reciprocal or nonreciprocal
- sister chromatids need not be identical
Technological limitations and challenges

- Biopsy
- Technology (FISH, WGA, SNP, NGS)
- Validation (problematic for ‘true mosaics’)
- Algorithms
- Resolution (segmentals)

(Fragouli et al., HRU, 2019)
(Capalbo and Rienzi, Fert Ster, 2017)
(Weissmann et al., Fert Ster, 2017)
(Harton, et al., Fert Ster, 2017)
(Vera-Rodriguez and Rubio, Fert Ster, 2017)
(Munne and Wells, Fert Ster, 2017)
(Marin, et al., Curr Opin Obstet Gynecol, 2017)
Origins of mosaic embryos

Meiosis (& PGC)  Embryonic mitosis  Tissue differentiation  Birth

sperm  sperm  zygote  cleavage stage  blastocyst  “black box”  fetus  births

mature MII  zygote  cleavage stage  blastocyst  fetus  births
Origins of mosaic embryos

Meiosis (& PGC) → Embryonic mitosis → Tissue differentiation → Birth

- **mature MII** → **zygote** → **cleavage stage** → **blastocyst**
- **70-90%** → **5-25%** → **1-2%** → **0.5%**

References:
- Vanneste et al., Nat Med, 2009
- van Echten-Arends, HRU, 2011
- Fragouli et al., HRU, 2019
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Origins and fates of mosaic embryos

Embryonic mitosis

$\text{cleavage stage} \quad \text{blastocyst}$

70-90% 5-25%

Maternal to zygotic transition (embryonic genome activation)
30-40% of embryos arrest
(markers of genome instability: yH2AX, micronuclei, DNA bridges, fragmented cells)
Common variants spanning PLK4 are associated with mitotic-origin aneuploidy in human embryos

Rajiv C. McCoy, Zachary Demko, Allison Ryan, Milea Banjevic, Matthew Hill, Styrmir Sigurjonsson, Matthew Rabinowitz, Hunter B. Fraser, Dmitri A. Petrov

Unstable maternal to zygotic transition

(McCoy et al., Science, 2015)
(McCoy et al., HMG, 2018)
(Zhang et al., Fert Ste, 2017)
Unstable maternal-zygotic transition: tripolar spindles?

HUMAN GENETICS

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ASSOCIATION STUDIES ARTICLE

Tripolar chromosome segregation drives the association between maternal genotype at variants spanning PLK4 and aneuploidy in human preimplantation embryos


Unstable maternal-zygotic transition: tripolar spindles?

Common variants spanning PLK4 are associated with mitotic-origin aneuploidy in human embryos

- Meiotic non-disjunction
- Mitotic anaphase lag
- Multiple aneuploidies

Aneuploidy

- Maternal chromosome gain
- Both maternal homologs detected in a single genomic region

Mechanism

Aneuploidy

- Maternal chromosome loss
- Paternal chromosome loss

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Tripolar chromosome segregation drives the association between maternal genotype at variants spanning PLK4 and aneuploidy in human preimplantation embryos

- ‘Chaotic karyotype’
- Day 3 embryos
- Arrest

(McCoy et al., Science, 2015)
(McCoy et al., PloS Genet, 2015)
(McCoy et al., HMG, 2018)
(Zhang et al., Fert Ste, 2017)
Mechanism leading to tripolar spindles

Dual-spindle formation in zygotes keeps parental genomes apart in early mammalian embryos

Judith Reichmann, Bianca Nijmeijer, M. Julius Hussain, Manuel Eguren, Isabell Schneider, Antonio Z. Politi, M. Julia Roberti, Lars Hufnagel, Takashi Hiiragi, Jan Ellenberg

…and when the two spindles fail to merge, embryos with ‘tripolar’-like spindles and multinucleated cells
Conservation in human embryos?

• Human embryos maintain separate pronuclei after fertilization
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- Spacial separation of parental genomes in human zygotes
  
  (van der Werken *et al.*, Nat. Comm., 2014)
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• Chaotic karyotypes result in cellular attrition and embryo arrest (Ottolini et al., 2017)
Basic research provides mechanistic insight

What are the mechanisms- and how many patients do they account for?
One gene, one therapy

Gene therapy for rare eye disease set to be offered on NHS

Patients with a rare inherited eye disorder are set to be able to access a new gene therapy on the NHS which slows down sight loss.

Draft NICE guidance recommends the therapy (Luxturna) to treat inherited retinal dystrophies.

The National Institute for Health and Care Excellence estimates just under 90 people in England will be eligible for the treatment.

Drug company Novartis agreed a discount for the NHS on the £613,410 price.

Without the deal, the therapy, voretigene neparvovec, would have breached the regulator's budget impact test.
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