

---

# Polycystic Ovary Disease: A Common Endocrine Disorder in Women

Paul Kaplan, M.D.

Clinical Professor of Reproductive Endocrinology - OHSU

Courtesy Senior Research Associate, Human Physiology

University of Oregon

# Case Presentation – Jenn A.

- 23 Y. O. G0 P0 menarche age 13
- BMI 29. Hx of “weight problems”.
- Menses Q 60 -180 days.
- Moderate acne & hirsutism age 14-15. Started BCPS age 15.
- Family Hx T2 diabetes and infertility



# Polycystic Ovary Syndrome

- Most Common Endocrine Disorder of R/A Women
  - Affects ~1/12 young women in U.S.(>10 Million)
  - Remains Underdiagnosed & Misunderstood
- Key Features:
  - Oligo/Amenorrhea
  - Abnormal Androgen Production & Metabolism
- Probable Genetic Etiology
  - ? autosomal dominant/variable penetration
  - Conveys evolutionary “metabolic efficiency”

# PCOS - History

---

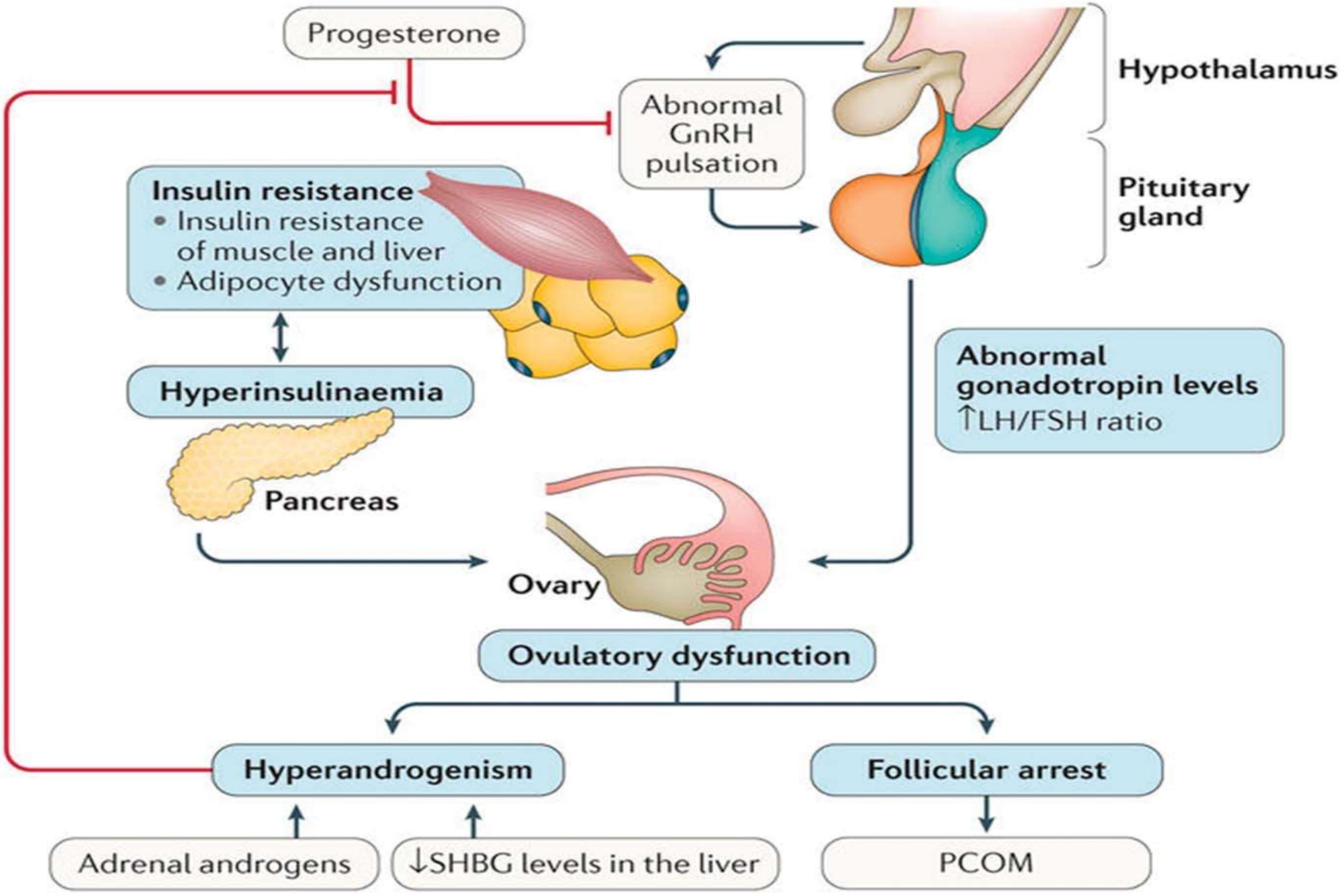
- 1935 “Stein-Leventhal Syndrome”
- Observed association of amenorrhea and polycystic ovaries (at surgery)
- Currently 30,000 published articles on PCOS
- Now recognized as the leading cause of infertility

# PCOS: A NEW PARADIGM

---

“ PCOS is a metabolic disorder affecting multiple body systems that requires comprehensive and long-term evaluation and management. ”

John Nestler, M.D. Fertility & Sterility November, 1998



# PCOS: Evolutionary Benefits

---

- Metabolic “Thriftiness”
  - Maximal caloric conservation
    - Reduced BMR
  - ↑ Longevity in animal models
  - Stress-induced ovulation ( ↓ LH P/F )
  - ↓ Rate of oocyte atresia ( ↑ Insulin levels )

# How Do Women with PCOS Present?

---

- ❑ Irregular Menstrual Periods
- ❑ Hirsutism
- ❑ Facial Acne
- ❑ Overweight/Obesity
- ❑ Infertility
- ❑ Acanthosis Nigricans (café au lait spots)





**Acanthosis Nigricans**

# PCOS: Diagnosis

- N.I.H. Definition (2 of 2)
  - Oligo/Anovulation
    - Cycles > 35 days apart or < 7 per year
  - Abnormal Androgen Production & Metabolism
    - Clinical (Hirsutism/Acne) or Lab (T, A, DHEA-S)
- ESHRE/ASRM Rotterdam 2003 (2 of 3)
  - Oligo/Anovulation
  - Androgen Excess
  - Polycystic Ovaries (20 or > follicles or >10cm<sup>3</sup>/ovary on U/S)
    - \* Genetics similar for both criteria (A. Dunaif 2019)

# PCOS

**YOUNG**

**LATER YEARS**

**REPRODUCTIVE  
AND  
HYPERANDROGENIC  
DYSFUNCTION**

**CARDIOMETABOLIC  
DISORDERS**

# PCOS: CVD Classification

- **Classic (75%)**
  - RD/NIH Criteria + Overweight (BMI > 25)
  - 40% Risk of IGT or T2DM by age 40 (5X controls)
  - Dyslipidemia in 70% (IR effect)
- **Ovulatory (Lean) (12.5%)**
  - Medium risk profile
- **Nonhyperandrogenic (12.5%)**
  - Lowest Risk

# PCOS: Clinical Consequences

---

- Endometrial Cancer (3x risk, up to 1/5)
- Spontaneous Abortion (? ↑ LH Effect)
- Gestational and Type 2 Diabetes (5-7x)
- Cardiovascular Disease (↑ LDL ↓ HDL)
- Hypertension
- Breast Cancer (3-4x risk in limited data)
- Ovarian Cancer
- Depression/Anxiety Disorders

# Evaluation of PCOS

---

- BMI, Waist Circumference, BP
- Baseline FSH, LH, TSH, Prolactin
- Testosterone, DHEA-S, AMH
- 17-OH Progesterone (Follicular a.m.)
- Fasting Glucose + Insulin/GTT
- Fasting Lipids & Chemistry Panel
- Transvaginal Ultrasound of Ovaries

RIGHT - 21 X 16

LEFT - 12 X 11

07/04/93  
09:07:00  
FROZEN  
FQ 75 75  
SC 06 06  
FC FM FM  
FR 10 10  
SS  
HR  
DL  
EE E2 E2  
DR 45 45  
CL 50 50  
TX-03 03  
RX 49 56

"String of Pearls" in PCOS



1H



\*WOMEN'S CARE FERTILITY CEN

# Transvaginal Ultrasound of the Ovaries

# PCOS: Insulin Resistance

---

- Demonstrated in 60 - 80% of PCOS Pts
  - 95% in Obese PCOS (BMI > 30)
- Metabolic Effects:
  - Decreased Hepatic SHBG Production (↑ Free T)
  - Increased Ovarian Thecal Androgen Production
  - Increased Triglycerides and Adverse Lipid Profile
  - Obesity/Metabolic Syndrome
  - Hypertension
  - High Risk of T2 DM (25%)



# Metabolic Syndrome: Diagnosis

---

- Three or more of the following:
  - Hypertension (130/85 or higher or on meds)
  - Elevated Triglycerides (>150 mg/dL or on meds)
  - Reduced HDL (Less than 50 mg/dL for women)
  - Waist circumference > 35 inches for women
  - Fasting Glucose >100 mg/dL or on meds

# Metabolic Syndrome: What We Know

- Occurs in 1/6 (16%) of the general population and 60% of obese men and women.
- 10% of people with NGT, 40% with IGT, 85% with T2 DM.
- Prevalence 24% higher in women (40% by age 60) and increases with age.
- Conveys a high risk of T2 DM and cardiovascular disease.
- Significant increased risk with PCOS
- Negative impact on fecundity and IVF outcome (JAMA 08/19)

# Metabolic Syndrome: Genetic Risk

- Mothers of PCOS Pts: RR 1.78 (95% CI 1.37-2.30)
- Sisters of PCOS Pts: RR 1.50 (1.12-2.00)
- Fathers of PCOS Pts: RR 1.43 (1.12-1.81)
- Similar Pattern seen in risk of Hypertension and Dyslipidemia

*Metabolic Syndrome, Hypertension, and Hyperlipidemia... in PCOS.  
Fert&Ster Feb2018;9109:2.*

# Metabolic Syndrome: Treatment

---

- Lifestyle: Diet, Exercise, Weight Loss
- Correction of Problems: HTN, DM, Lipids
- High-risk monitoring/follow-up

# PCOS: Cardiovascular Disease

---

- Dyslipidemia
- Hypertension
- Impaired Glucose Tolerance/Type2DM
- Metabolic Syndrome
- Frequent Positive FH CVD before age 55
- Carotid-IMT (10-15% over controls)
- Carotid Artery Calcification
- Multivessel CVD (32% vs. 25%)

# PCOS: Treatment Options

---

- Anovulation: Cyclic Progestins, BCPs
  - Prevent D.U.B., Endometrial CA
- Acne/Hirsutism: BCPs, Spironolactone
- Contraception: Low-Androgenic BCPS
- Fertility: Aromatase Inhibitors, Clomiphene, FSH/hMG, IVF
- Weight Loss: Low Calorie ADA Diet
- *Role of Insulin-Sensitizing Medications*

# PCOS: Positive Effects of Insulin Sensitizing Agents

---

- ↑ SHBG ( ↑ Androgen Binding)
- ↓ Testosterone and Androstenedione
- ↓ Triglycerides and LDL
- Regulation of Menstrual Cycles (30%)
- Weight Loss (Slow)
- Increased Sensitivity to Ovulation Meds
- ? Decreased Risk of Miscarriage

# PCOS: Who to Treat with DM Meds in 2020

---

- No current FDA approved therapies for PCOS
- Metformin data strongest to date
  - Thiazolidinediones, GLP-1, DPP-IV, SGLT2 have role
- Obese (BMI>30) PCOS patients
  - Adults & Adolescents (who can be compliant)
- Insulin resistant patients
  - Fasting Insulin Elevated or G/I ratio < 4.5
- Young patients (age <30) attempting pregnancy
- Patients with impaired glucose tolerance (IGT) or Type 2 D.M.



# PCOS: Management Summary

---

- Tailor treatment to life stage
  - Stress healthy diet/regular exercise
- Induce regular menses
- Identify & treat endocrinopathies
- Identify & treat insulin resistance
- Ongoing regular medical follow-up

# CLOSE TO HOME



**With prescription drug prices rising exponentially, many drugstores now provide armed escorts to assure that customers reach their cars safely.**

# Irving Stein, M.D.



# Michael Leventhal, M.D.



**Assisted Reproductive**  
**Technologies: Present**  
**and Future**

Paul Kaplan, M.D.

# Case Presentation – ART

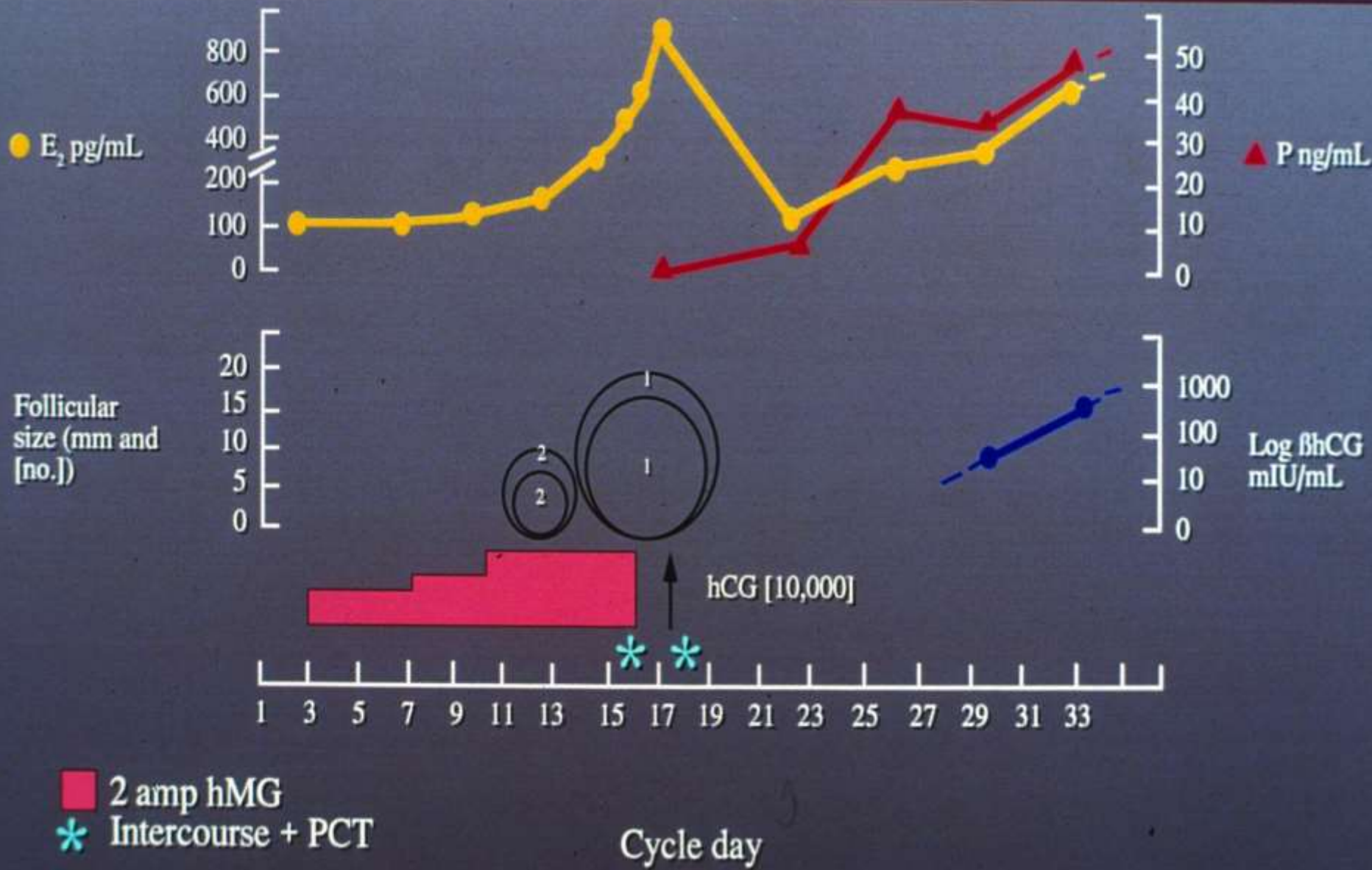
- 38 Y. O. Female G0 P0 Biochemist
- 40 Y. O. Male Software Engineer
- Infertility x 4 years
- Negative Work-up
- Unsuccessful ovulation induction +  
I.U.I x 6



# The Assisted Reproductive Technologies (ART)

- **In Vitro Fertilization (IVF)**
- **Intracytoplasmic Sperm Injection (IVF/ICSI)**
- **Donor Oocyte IVF**
- **Frozen Embryo Thaw and Transfer**
- **Cryopreservation/In Vitro Maturation of Oocytes**

# An hMG-hCG Cycle



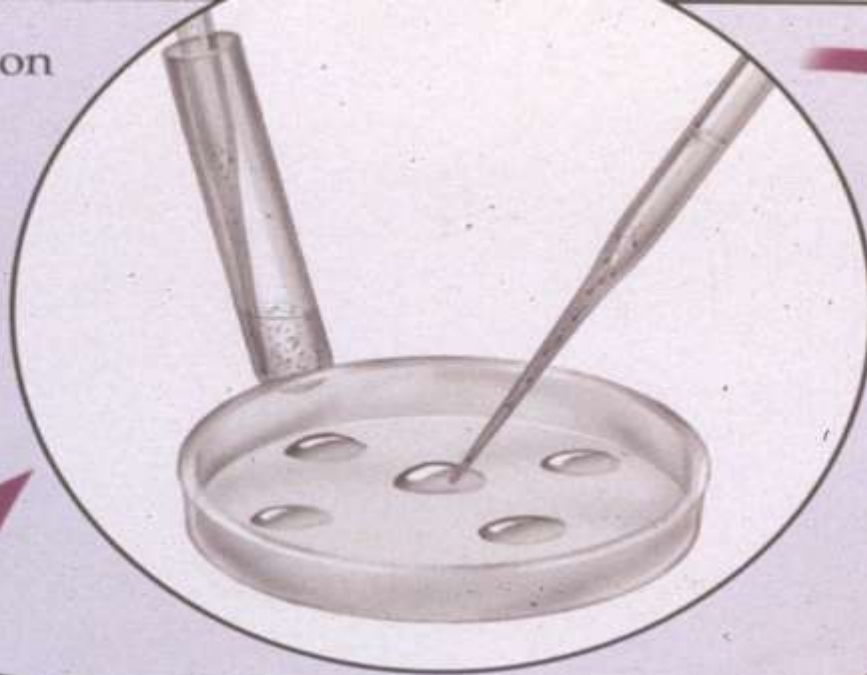
Adapted from Navot and Rosenwaks, 1987.



# In Vitro Fertilization (IVF)

- Daily S/C or IM FSH/hMG injection
- Follicular monitoring with serum estradiol and transvaginal ultrasound
- HCG given to trigger ovulation (LH surge)
- Transvaginal oocyte retrieval and insemination
- Embryo culture and transcervical embryo transfer
- Embryo cryopreservation for future F.E.T.
- Pregnancy rate of 40-50 % per cycle

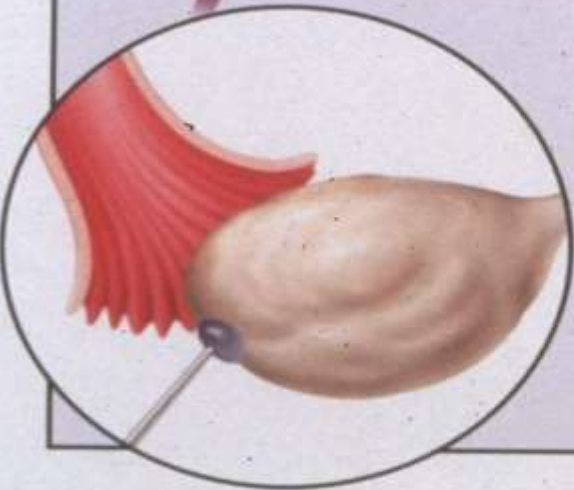
Insemination



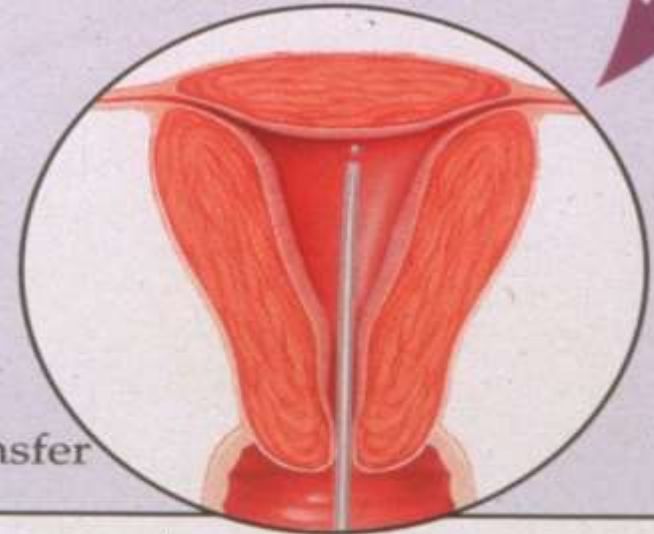
Incubation



Egg Aspiration



Embryo Transfer



OTTC

In IVF, eggs are harvested from the woman's ovary and fertilized in the laboratory with sperm. The embryos are then transferred into the uterus.

## IVF Embryo Culture and Transfer

# Intracytoplasmic Sperm Injection (ICSI)

- Standard IVF Stimulation and oocyte retrieval
- Injection of a single sperm into each oocyte
- Embryo culture and transcervical embryo transfer
- Currently used in almost 50% of IVF cycles for treatment of male factor and unexplained causes
- Pregnancy rate of 40-50 % per cycle

# Intracytoplasmic Sperm Injection (ICSI)



# *Current Directions in ART*

- The “*-omics*” revolution in non-invasive screening:
  - Genomics, Proteomics, Metabolomics, Embryomics
- Preimplantation genetic diagnosis (PGD)
  - *with gene therapy?*
- Nuclear and/or cytoplasmic oocyte transfer
- Embryonic Stem Cell Line Development
- Gamete Stem Cell Development

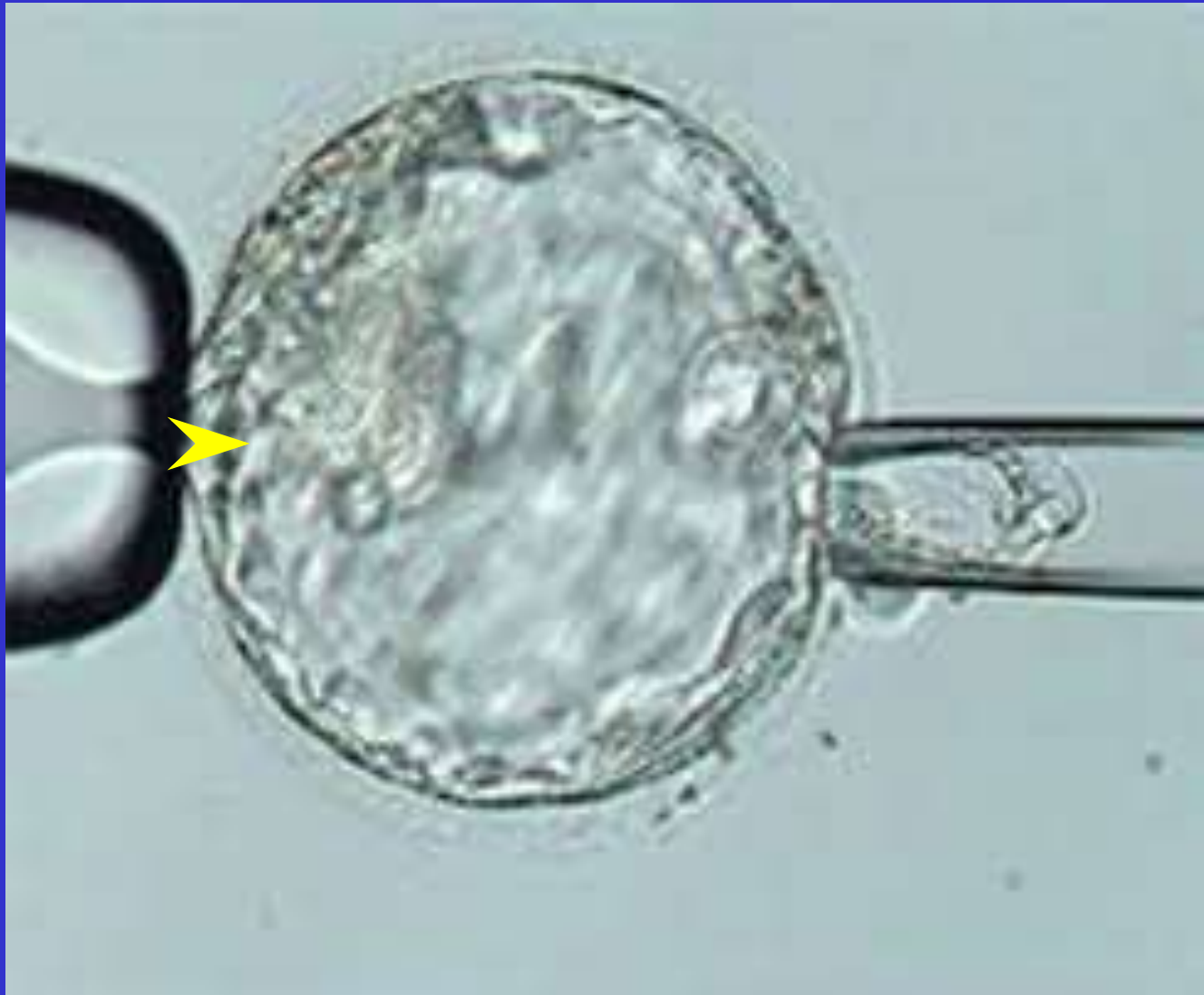
# *Future Directions in ART ??*

- Embryo Cloning - Reproductive/Therapeutic
- Adult Stem Cell Line Creation
- Adult Cell Gamete Cloning - sperm/oocyte
- Adult Somatic Cell Cloning

# *Preimplantation Genetic Diagnosis (PGD)*

- Goal: Identify Genetically Abnormal Embryos
- IVF/ICSI + Embryo Culture
- Trophectoderm Biopsy of Blastocyst (~day 5)
- CCS (comprehensive chromosomal screening)
- Transfer of Normal Blastocysts/Frozen Embryos
- Next Steps ??

# PGD Trophectoderm Blastocyst Biopsy



ICM



# *Comprehensive Chromosomal Screening (CCS)*

- **Next Generation Sequencing (NGS)**
- **Quantitative PCR (qPCR)**
- **Single Nucleotide Polymorphism (SNP array)**
- **DNA Fingerprinting**
- **Comparative Genomic Hybridization (array CGH)**
- ***Whole Genome Sequencing (future)***

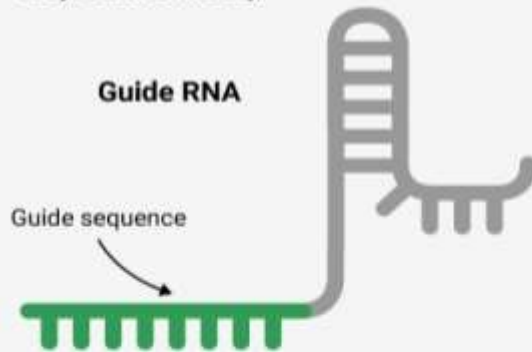
## How Will CRISPR Technology change PGD ??

- *Clustered Regularly Interspaced Short Palindromic Repeats*
- Rapidly remove/add genes using Cas nuclease and synthetic guide RNA
- Has been successfully used to modify human hematopoietic cells in sickle cell disease
- Major potential for embryo gene defect correction
- Ongoing human research in China, UK, and US on nonviable & viable embryos
- Huge ethical issues ??

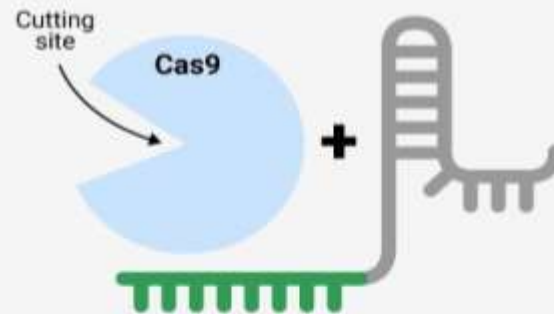
# CRISPR Technology

## EDITING A GENE USING THE CRISPR/CAS9 TECHNIQUE

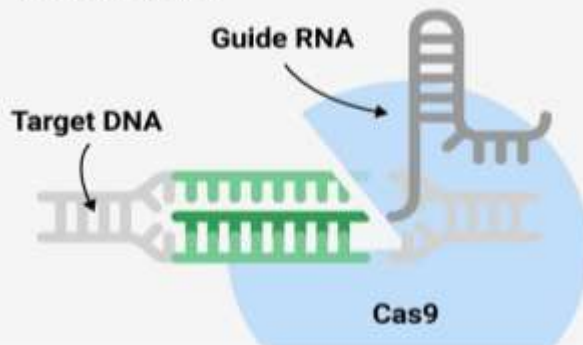
- 1** Scientists create a genetic sequence, called a "guide RNA," that matches the piece of DNA they want to modify.



- 2** This sequence is added to a cell along with a protein called Cas9, which acts like a pair of scissors that cut DNA.



- 3** The guide RNA homes in on the target DNA sequence, and Cas9 cuts it out. Once their job is complete, the guide RNA and Cas9 leave the scene.



- 4** Now, another piece of DNA is swapped into the place of the old DNA, and enzymes repair the cuts. Voilà, you've edited the DNA!



# Gene editing for Congenital Hypertrophic Cardiomyopathy



KRISTYNA WENTZ-GRAFF/Oregon Health & Science University

**Oregon Health & Science University** researcher Shoukhrat Mitalipov (left) talks with research assistant Hayley Darby in the Mitalipov Lab at OHSU in Portland.

## Embryo gene-repair holds promise for inherited disease

*A new journal article details the work at Oregon's medical school, which targeted a heart defect*

**BY LAURAN NEERGAARD**  
The Associated Press

line" changes — altering sperm, eggs or embryos — are controversial because they would be permanent, passed down to future generations. Critics worry about attempts at "designer babies" instead of just preventing disease, and a few previous attempts at learning to edit em-

**Nature 548:7665**  
**August 2017**

## Scientist announces gene-edited babies

By Lauran Neergaard  
and Malcolm Ritter

The Associated Press

Designer babies might be here sooner than anyone reckoned. A Chinese researcher who says he created gene-edited babies crossed what most scientists consider a forbidden line.

It's not clear if the claim is true and if so, how the twin girls whose DNA reportedly was altered will fare as they grow.

There is wide scientific agreement that rewriting DNA before birth — to prevent an inherited disease or to give a baby some “designer” trait — isn't yet safe to try outside laboratory experiments

that do not lead to human births.

“Grossly premature and deeply unethical,” is how noted U.S. bioethicist Henry Greely of Stanford University characterized the claim.

The researcher, He Jiankui of Shenzhen, said he altered embryos when parents were undergoing fertility treatments to change a gene so that it might provide the resulting babies with a trait few people naturally have — protection against future infection with the AIDS virus.

“This is probably the worst gene you would choose” to test in pregnancy because it doesn't fix a disease the children

were destined to get, said Shoukhrat Mitalipov of the Oregon Health & Science University, who in laboratory-only experiments studies how to repair gene defects in embryos.

“Where is the assurance this mutation now will result in resistance to HIV?” Mitalipov added. “He's testing his hypothesis on babies.”

Here are questions and answers about Monday's claim and the state of gene editing:

### What is gene editing?

It's a technology that lets scientists alter the DNA of living cells — from plants, animals, even

humans — more precisely than ever before. It's like a biological cut-and-paste program.

### How is it used?

Researchers routinely use gene-editing tools in labs to study diseases in cells or animals, and they're altering crops and food animals for better agriculture.

But in people, gene editing still is highly experimental. One first-in-human study is testing intravenous infusion of gene-editing ingredients to fight a killer metabolic disease. Other researchers are developing ways to gene-edit damaged cells and return them,

repaired, into patients with sickle cell disease and other disorders. But unlike Monday's announcement, none of those experiments would alter DNA in a way that patients would pass to their own children.

### What did the Chinese scientist do?

The researcher said he used the gene-editing tool CRISPR to alter a gene named CCR5 in embryos for seven couples during their fertility treatments; one pregnancy resulted. A particular mutation in that CCR5 gene is thought to confer some resistance to HIV by making it harder for that virus to enter cells.

### Why is the news so controversial?

Altering genes in sperm, eggs or embryos means those changes can be passed down to future generations — people who would have no way to consent to those changes. Plus, long-term negative effects might not become apparent for years.

In 2017, the U.S. National Academies of Science, Engineering and Medicine said lab-only research to learn how to alter embryos is ethical — but said it's not ready for pregnancies yet. The academy said if it is ever allowed, it should be reserved to treat or prevent serious diseases with no good alternatives.

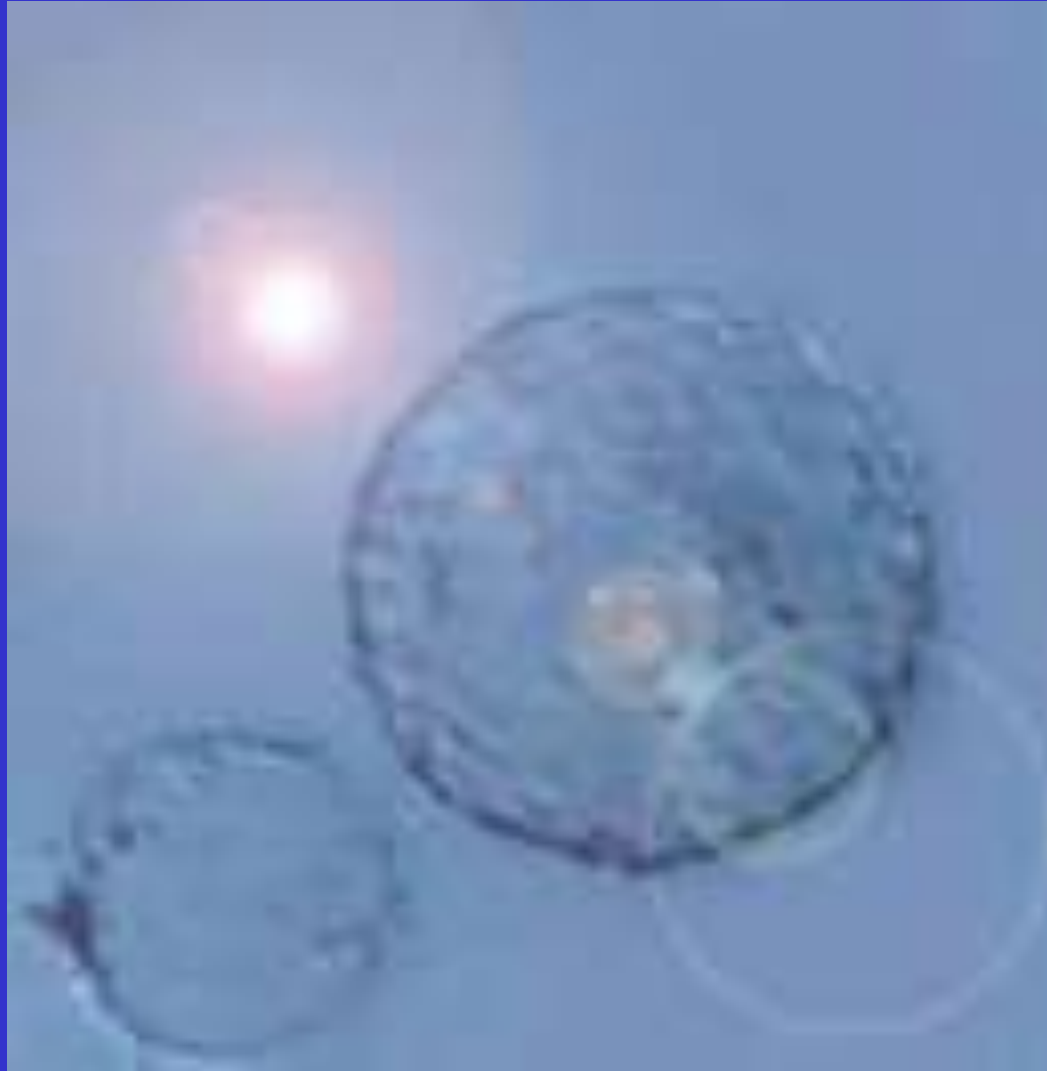
# Gene Editing in IVF

- Dr. He Jiankui (Shanghai, China) announced birth of IVF twins 11/18
- Used CRISPER/Cas to modify genes for HIV resistance
  - Father HIV Positive
- Widely criticized for lack of oversight
- Both babies healthy so far
- 2<sup>nd</sup> ongoing pregnancy announced in program
- Met with international condemnation due to unknown risks of genomic mutation and mosaicism

# Oocyte Cryopreservation

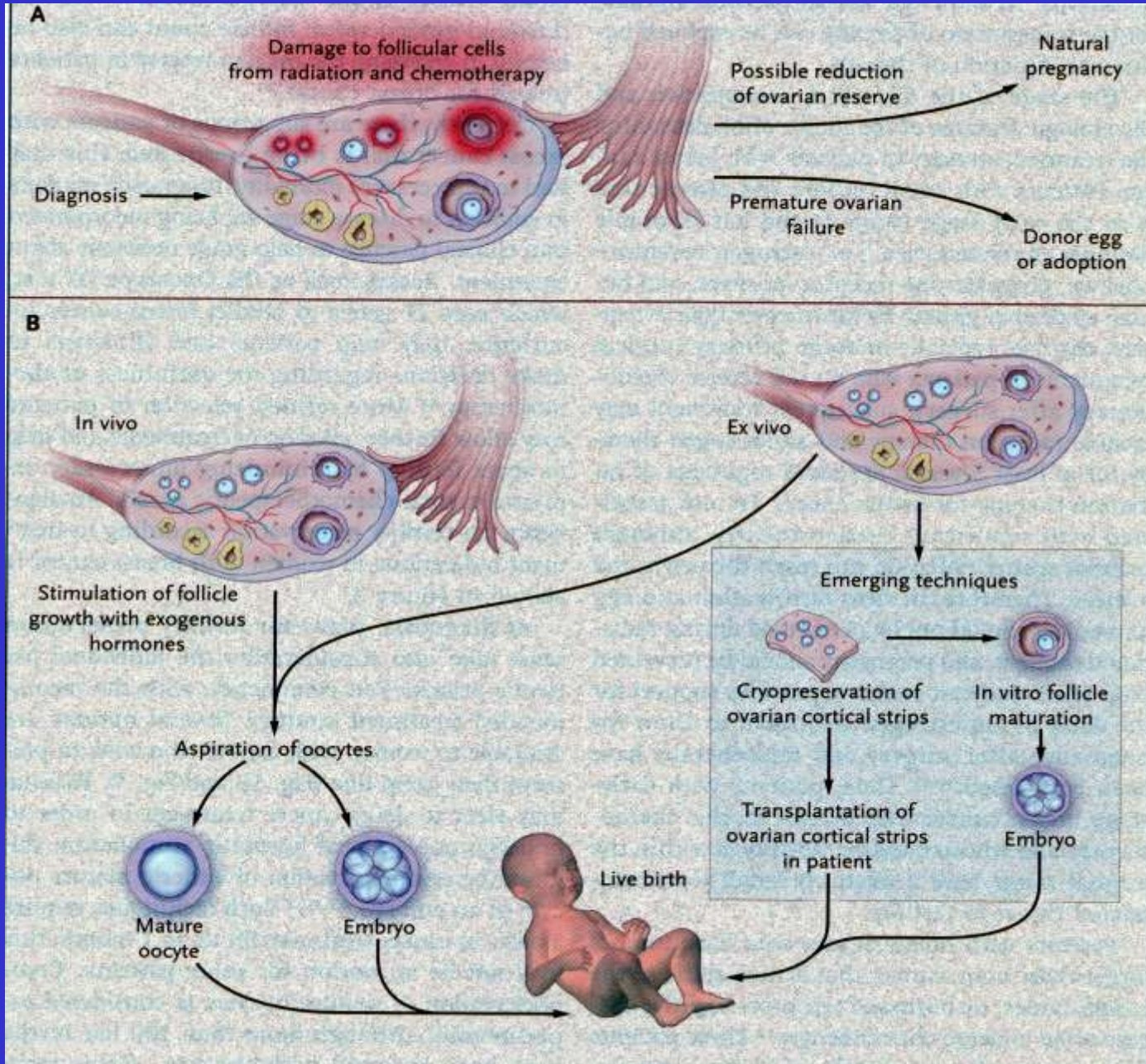
- Preservation of Oocytes Prior to Fertilization
- TV Retrieval of Stimulated Oocytes
  - Future: Unstimulated Oocytes with IVM
- Desiccation and Cryopreservation
- Delayed Thaw and IVF/ICSI Embryo Culture
- Transfer of Healthy Embryos

# Oocyte Desiccation for Cryopreservation

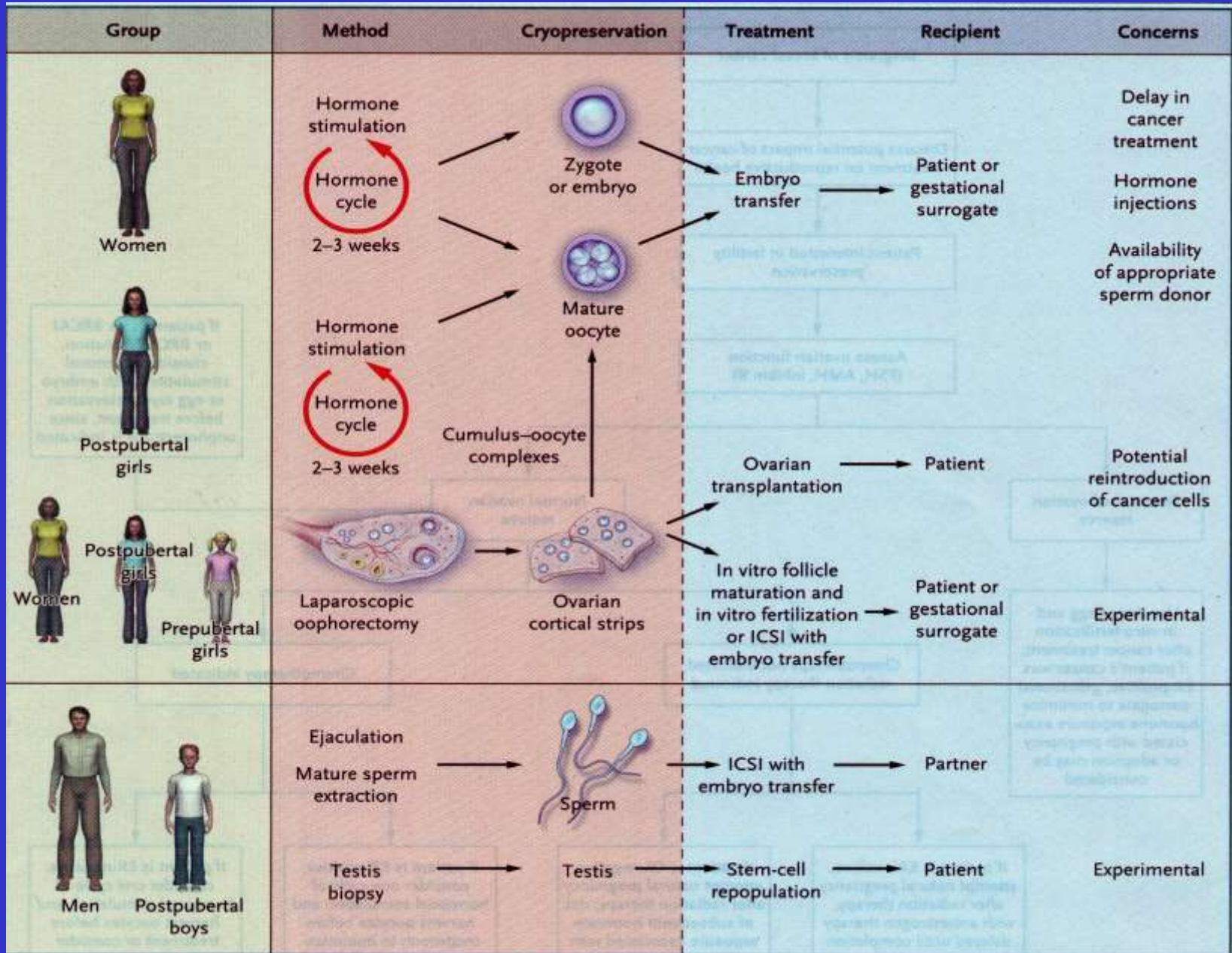




# Fertility Preservation



# Fertility Preservation



# IVF Monkey From Prepubertal Testis Bx



**“Grady” – OHSU Primate Center 03/2019**

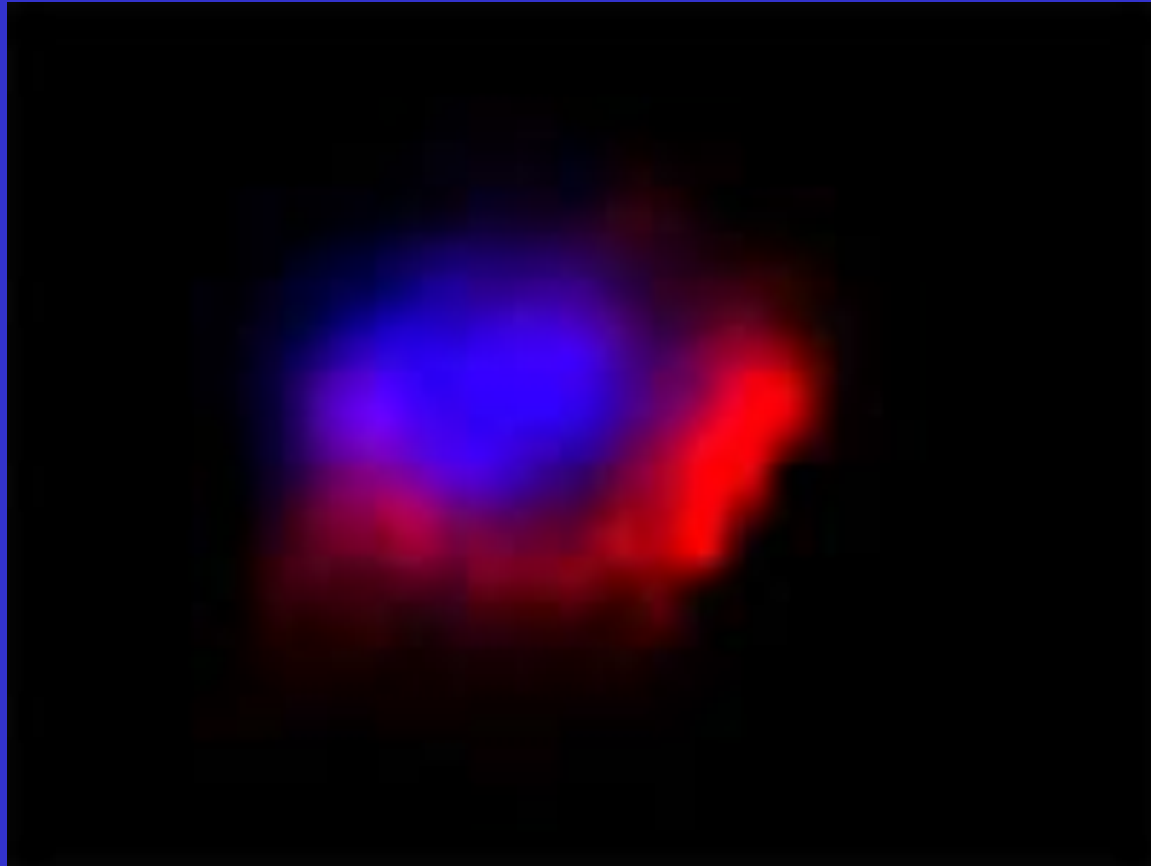
# IVF Treatment of Genetic Mitochondrial Disease

- **“Mitochondrial DNA Replacement Therapy”**
  - Third Party Reproduction
- **Performed by Nuclear Transfer to Donor Egg**
- **1:200 incidence of pathogenic mtDNA mutations**
- **UK: Several Ongoing Pregnancies**
- **One Live Birth (Leigh syndrome) in Mexico (U.S. Team)**
- **Currently in active IRB-approved study at OHSU**

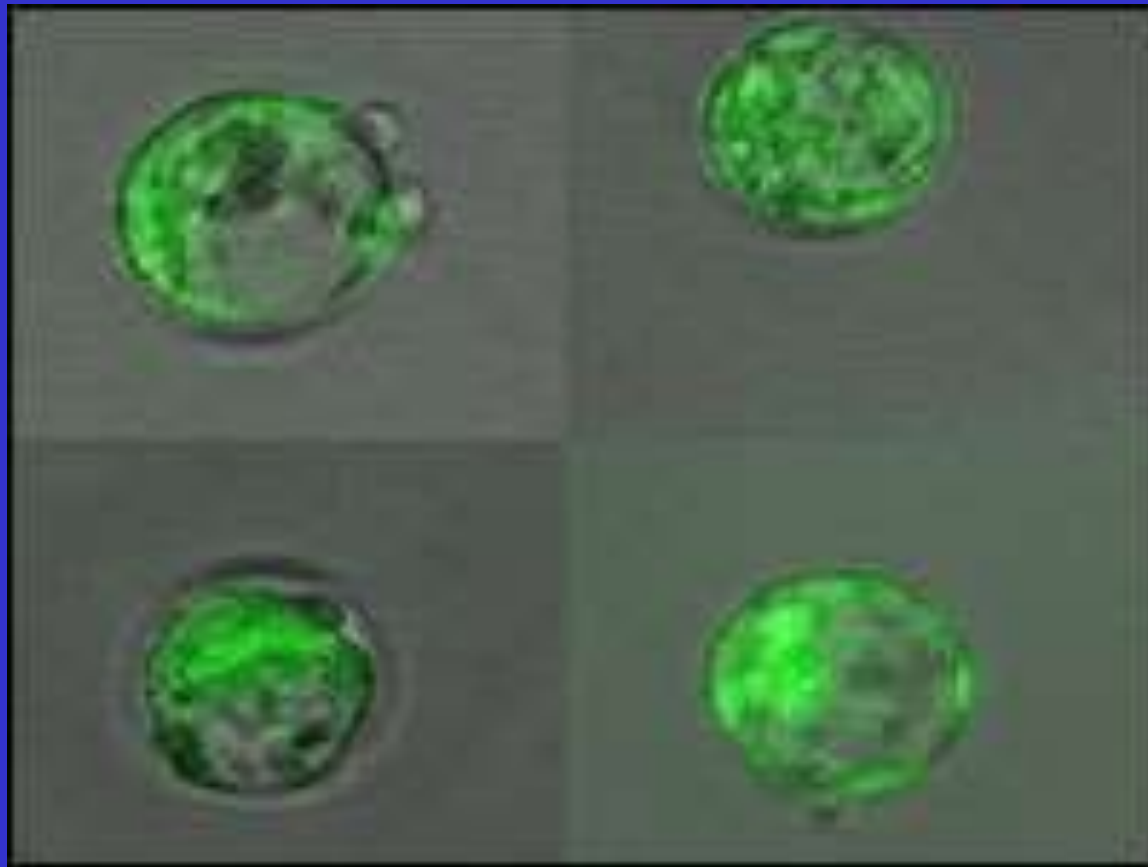
# Stem Cell Gamete Production

- A. Goal is cost-effective, ethically-acceptable source of sperm and oocytes**
  - Reduction of risks with donor gametes
  - Alternative to somatic cell cloning
- B. Potential treatment for cancer patients, age-related infertility, and severe male factor**
- C. Same sex couple biologic offspring**
- D. Reduction of multiple gestation by SET**

# Mouse Embryonic Stem Cell Spermatid



# Mouse Blastocysts Fertilized by Embryonic Stem Cell Spermatids



# Same Sex Couple ART

- A. Embryonic stem cells from a female mouse**
- B. Delete maternal imprinting genes with Crispr**
- C. ICSI modified cells into eggs of another female**
- D. Healthy mouse pups developed**
- E. Not yet successful with male mice**



# Mice With Two Moms



The Week Nov. 2018

# Milestones of infertility medicine



**1969**

Pergonal and human chorionic gonadotropin marketed

**1981**

First IVF baby in America

**1985**

Maryland passes legislation requiring insurance coverage for IVF

First ultrasound-guided, nonsurgical IVF

**1990**

Mark Sauer reports pregnancies in postmenopausal women

**1987**

ZIFT technique introduced

Lupron comes on the market

**1992**

Fertility Clinic Success Rate and Certification Act calls for uniform definition of success; to take effect October 1994

**1967**

Clomid comes on the market

**1978**

Louise Brown, first "test-tube baby," born

**1984**

GIFT technique developed by Ricardo Asch of San Antonio

First "donor baby" (eggs and sperm) born to surrogate mother in Australia

**1986**

Richard Marrs delivers first U.S. baby developed from a frozen embryo

**1991**

First preimplantation genetic screening (for cystic fibrosis)

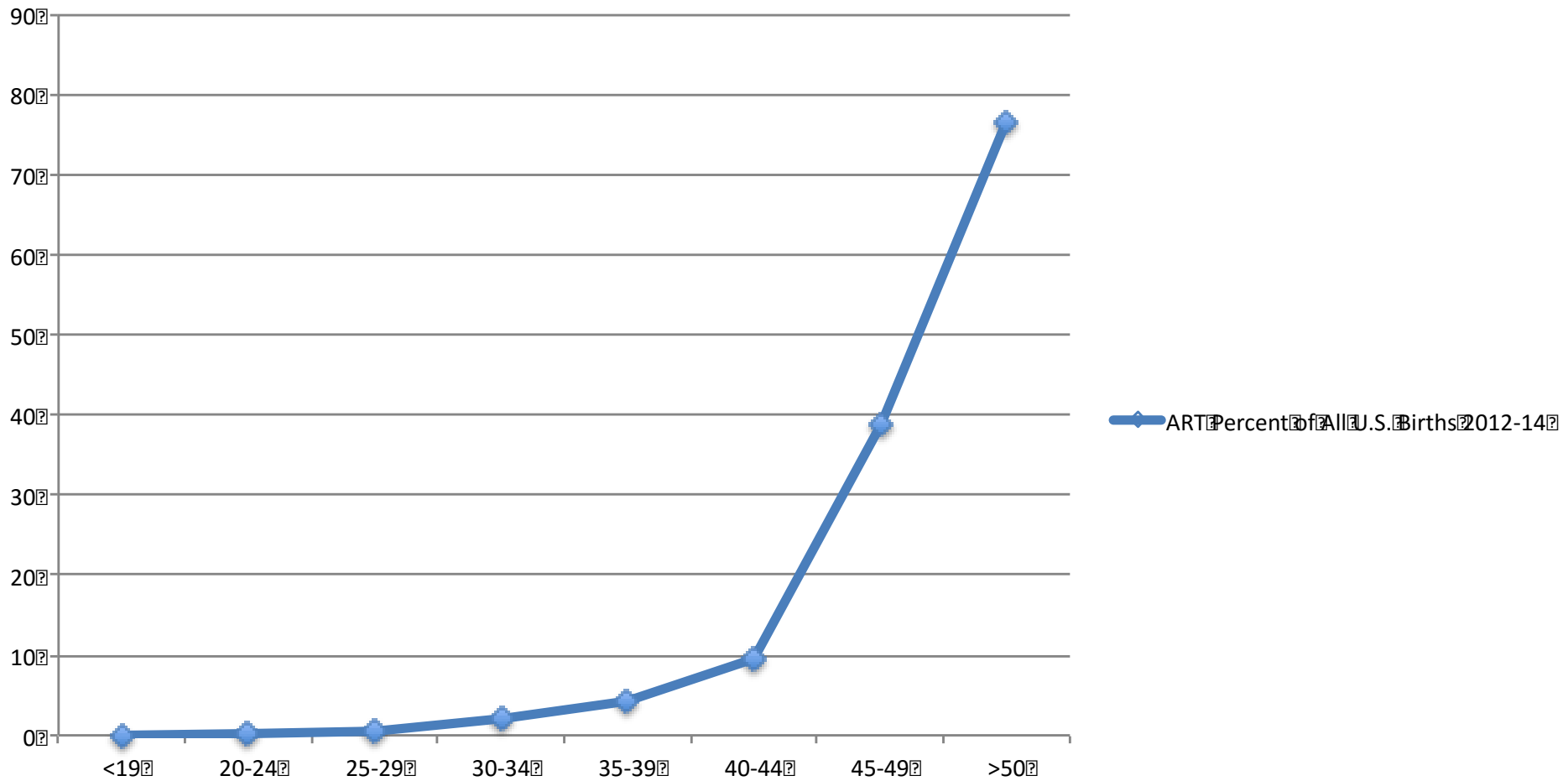
**1993**

Supreme Court decides frozen embryos cannot be implanted against the father's will

# In Vitro Fertilization (IVF) - 2020

- SART Data: 78,052 IVF babies born in 2017 in U.S.
- IVF babies now constitute almost 2% of U.S. births
- Estimated 650,000 IVF babies born in 2017 in world
- IVF births now almost 4% of births in Europe
- Estimated >8,000,000 IVF births WW by Jan. 2018
  - Who Knew ?????

## ART Percent of All U.S. Births 2012-14





© AFP/Getty Images

The world's first IVF baby Louise Brown (2nd right) posing with her son Cameron, her mother Lesley Brown and IVF pioneer Professor Robert Edwards in 2008

# OHSU IVF Team Reunion Aug. 2017



# How To Make A Vegan.....

