TURNING SKIN CELLS INTO GAMETES

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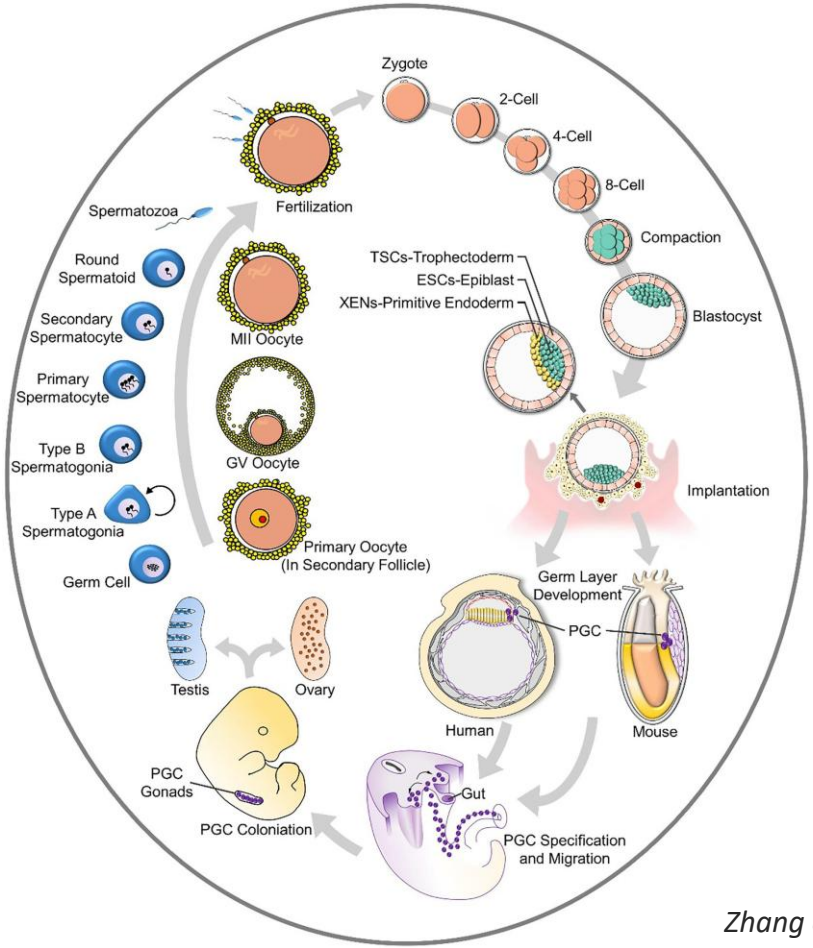
Why to generate gametes from somatic cells?

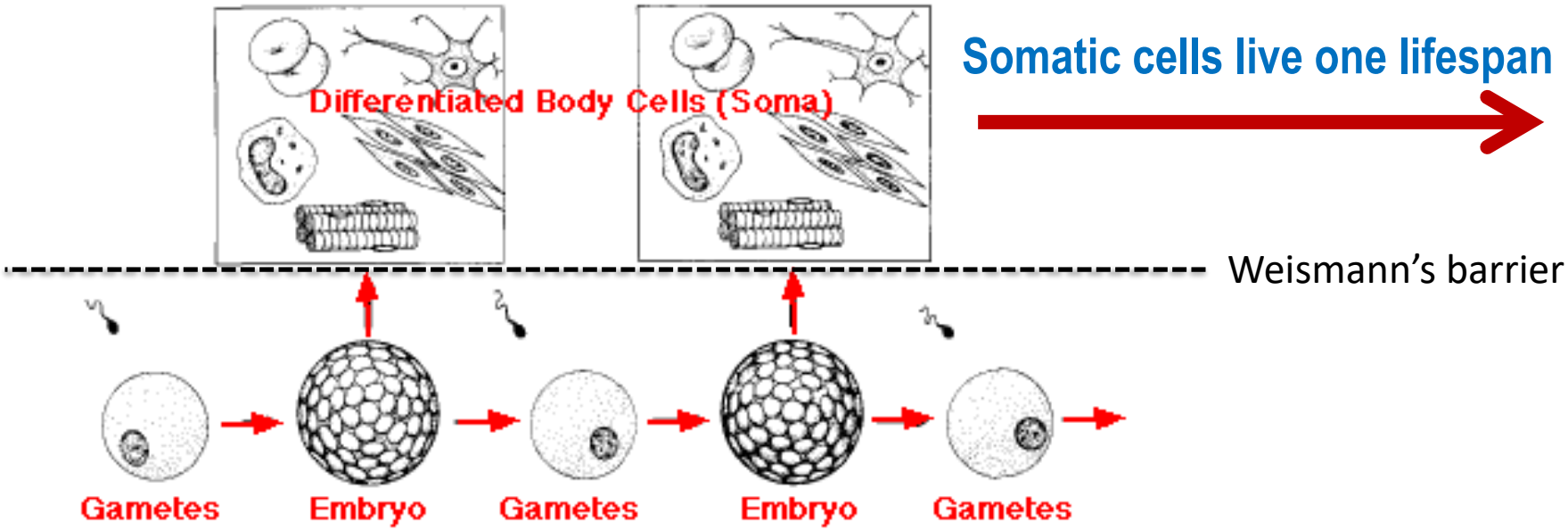
- Infertility treatment for patients who are unable to produce their own functional gametes thus, allowing them to have genetically related children
  - ✓ Advanced age
  - ✓ Cancer treatments
  - ✓ Genetic disease
  - ✓ Other medical conditions or injury
  - ✓ Same-sex couples
- Research
- Preservation

Cycle of Embryo and Gamete Development (Germline)

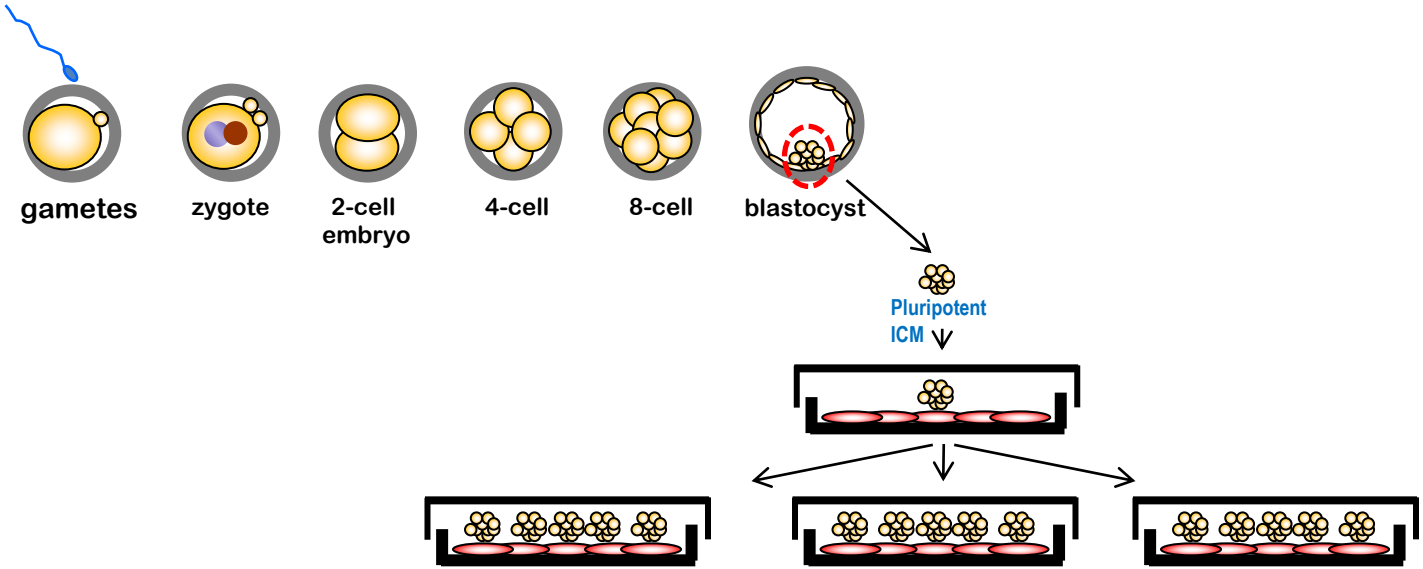


Zhang et al, Front. Bioeng. Biotechnol, 2020

August Weismann's Theory of the continuity of germplasm (1892)

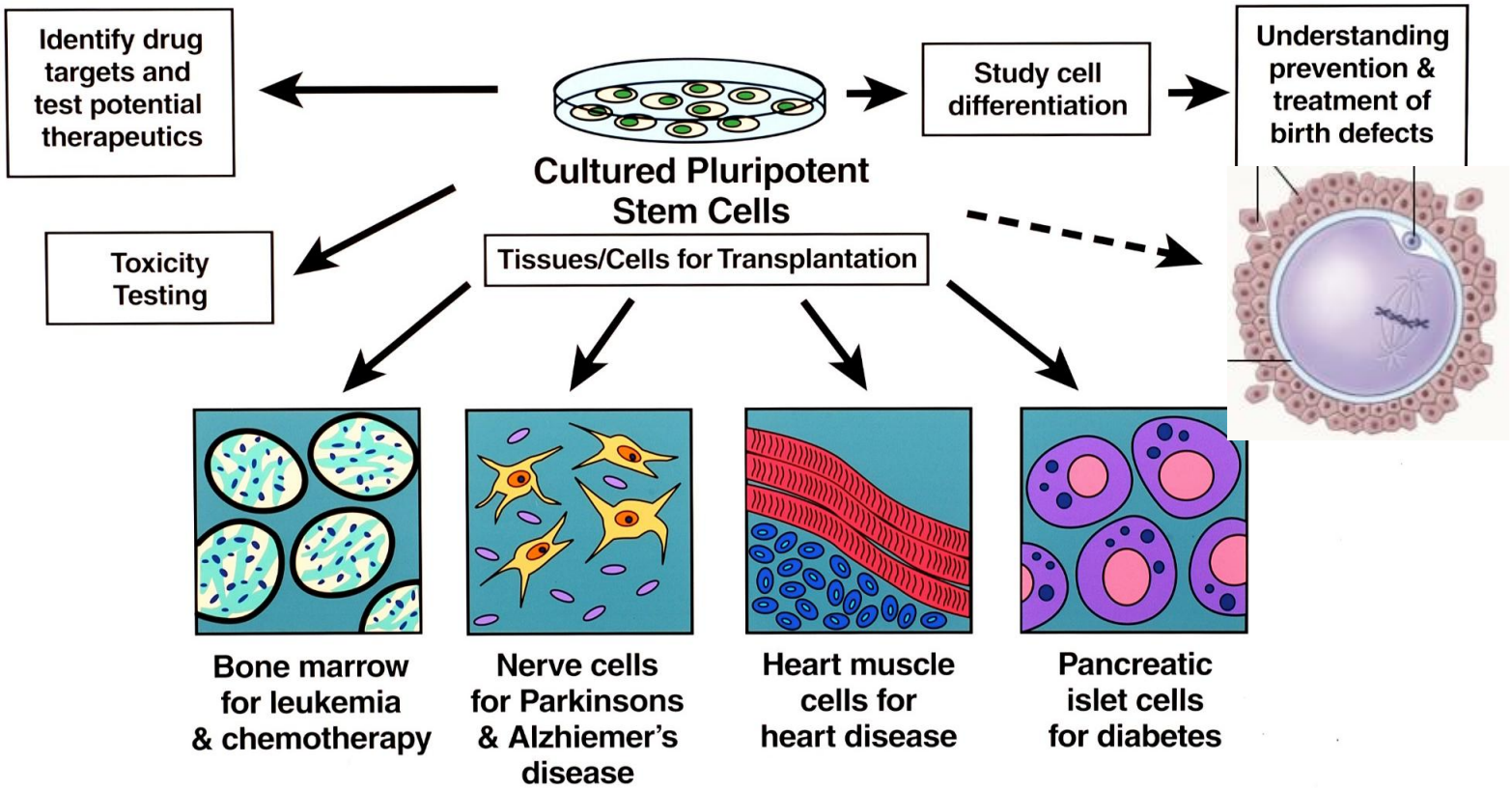


Embryonic stem cells (ESCs)

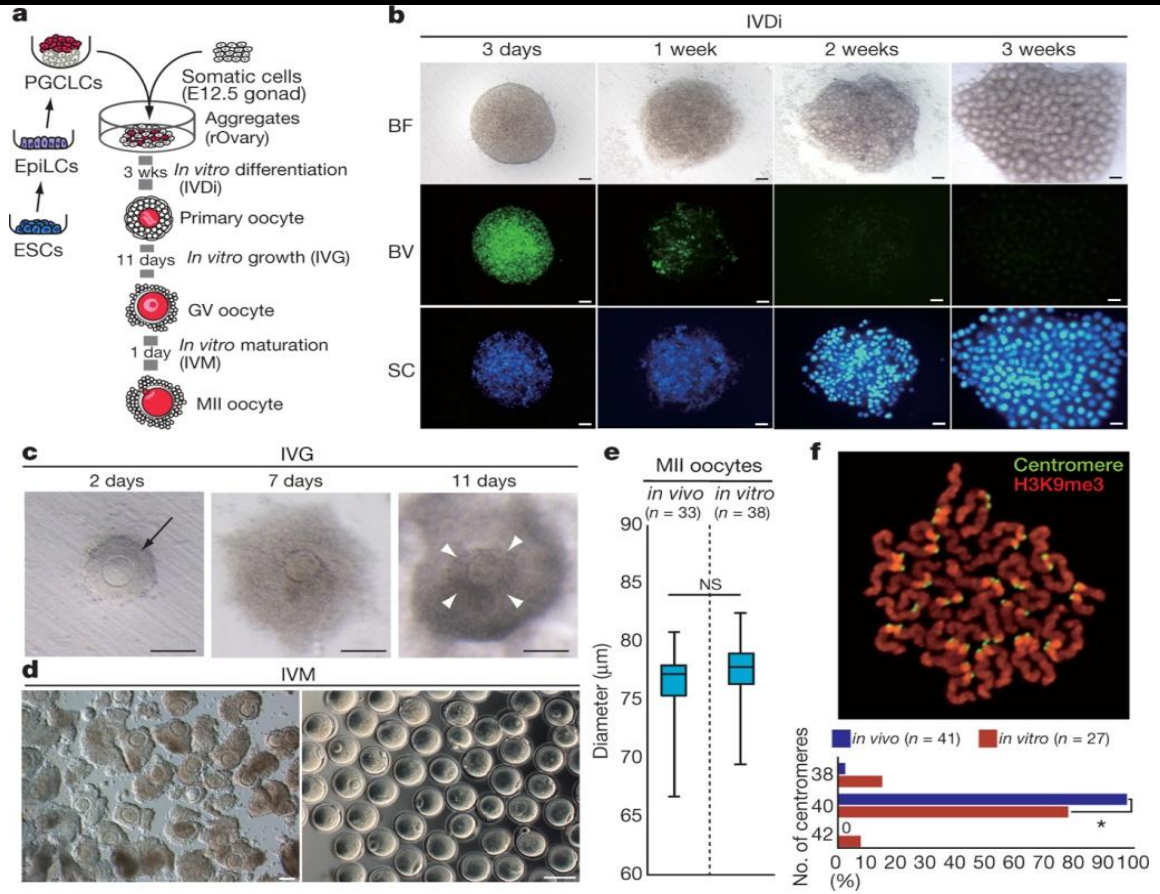


Pluripotent ESCs

ESCs for Regenerative Medicine

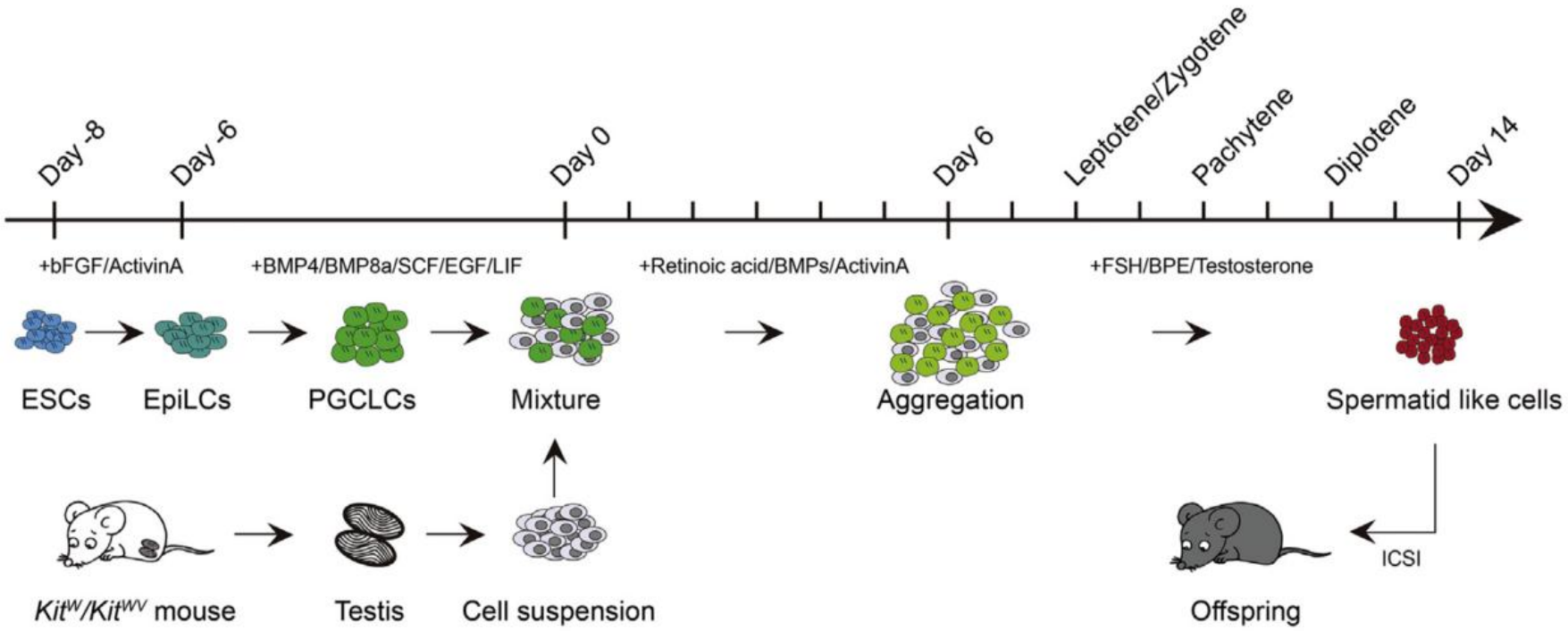


Mouse oocytes derived from ESCs



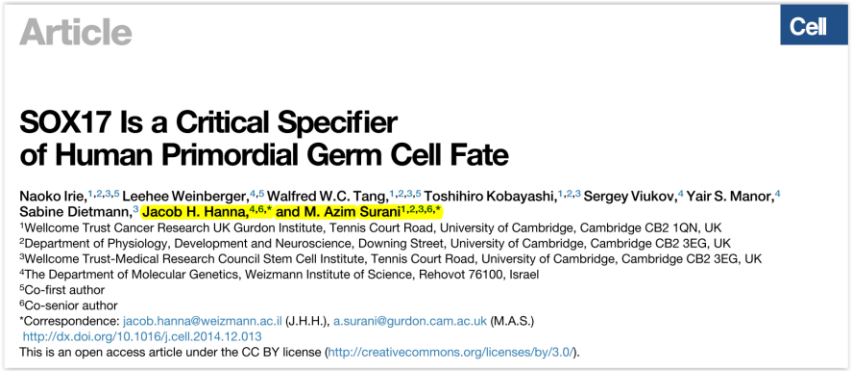
Hikabe *et al. Nature* (2016)

Mouse sperm produced from ESCs



Zhou et al. Cell Stem Cell. 2016

Human gametes from ESCs?



Article

Cell

SOX17 Is a Critical Specifier of Human Primordial Germ Cell Fate

Naoko Irie,[1,2,3,5] Leehee Weinberger,[4,5] Walfred W.C. Tang,[1,2,3,5] Toshihiro Kobayashi,[1,2,3] Sergey Viukov,[4] Yair S. Manor,[4] Sabine Dietmann,[3] Jacob H. Hanna,[4,6,*] and M. Azim Surani[1,2,3,6,*]

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[2]Department of Physiology, Development and Neuroscience, Downing Street, University of Cambridge, Cambridge CB2 3EG, UK
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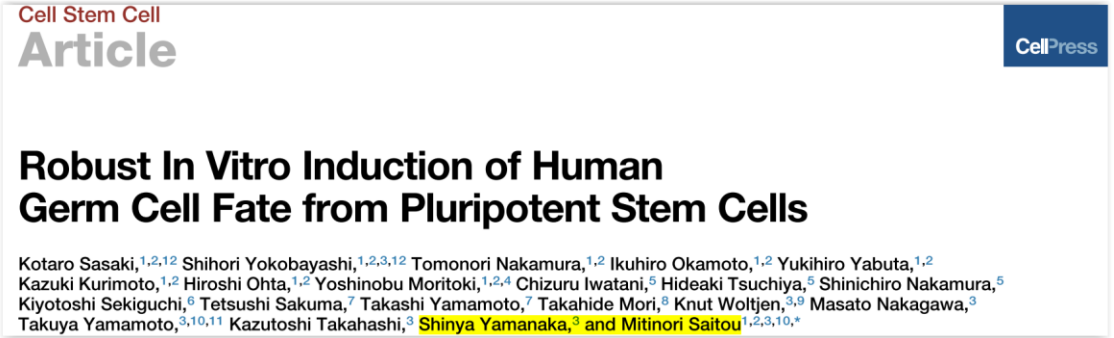
http://dx.doi.org/10.1016/j.cell.2014.12.013



Cell Stem Cell

Article

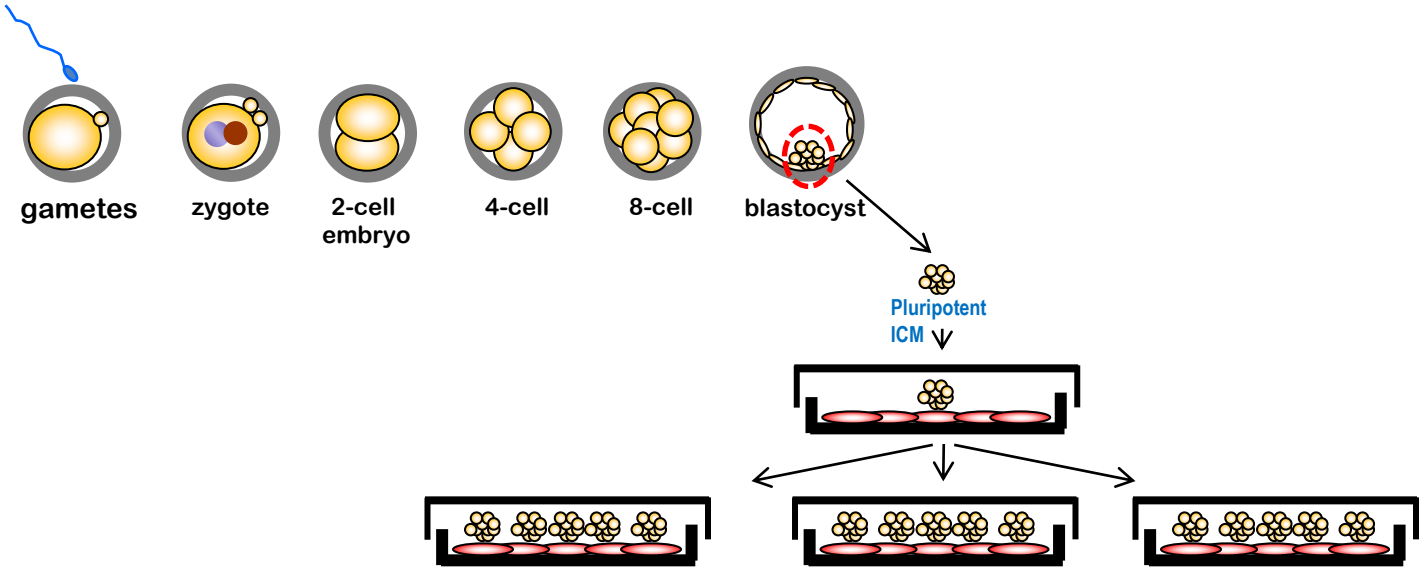
CellPress

Robust In Vitro Induction of Human Germ Cell Fate from Pluripotent Stem Cells

Kotaro Sasaki,[1,2,12] Shihori Yokobayashi,[1,2,3,12] Tomonori Nakamura,[1,2] Ikuhiro Okamoto,[1,2] Yukihiro Yabuta,[1,2] Kazuki Kurimoto,[1,2] Hiroshi Ohta,[1,2] Yoshinobu Moritoki,[1,2,4] Chizuru Iwatani,[5] Hideaki Tsuchiya,[5] Shinichiro Nakamura,[5] Kiyotoshi Sekiguchi,[6] Tetsushi Sakuma,[7] Takashi Yamamoto,[7] Takahide Mori,[8] Knut Woltjen,[3,9] Masato Nakagawa,[3] Takuya Yamamoto,[3,10,11] Kazutoshi Takahashi,[3] Shinya Yamanaka,[3] and Mitinori Saitou[1,2,3,10,*]

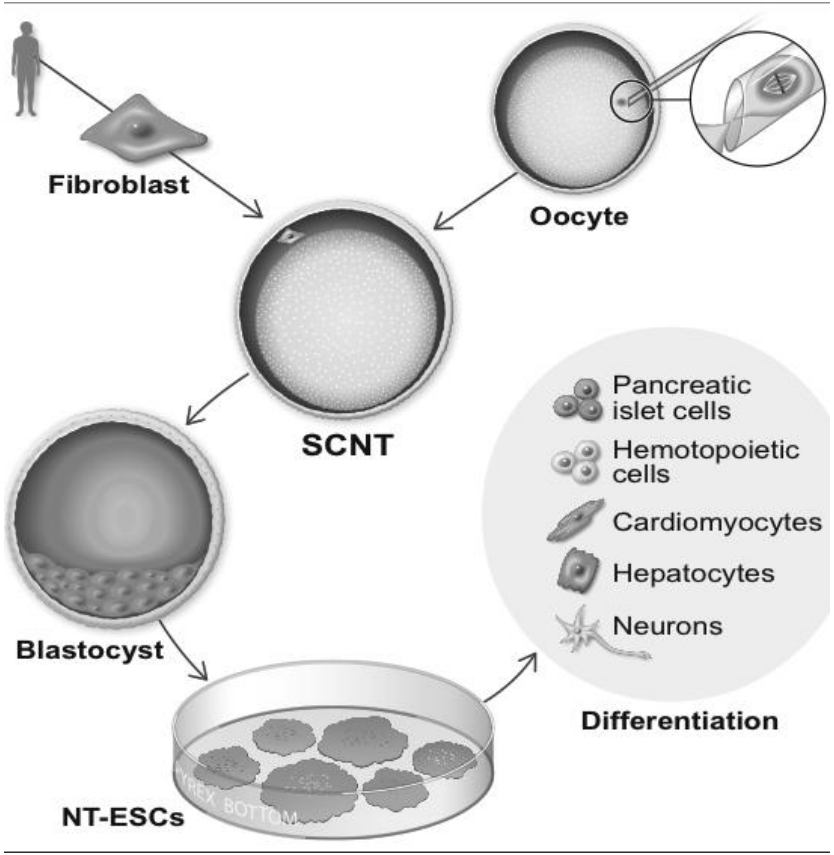
Reconstitution of Folliculogenesis In Vitro: Meiosis Is the Biggest Obstacle

ESCs cannot be produced for patients that lack functional gametes



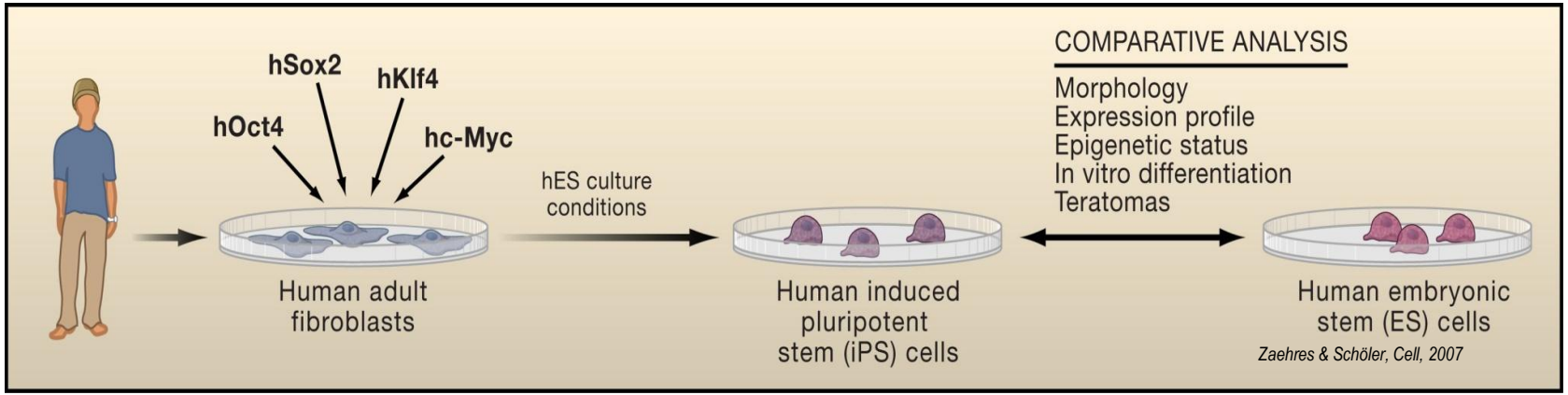
Pluripotent ESCs

Human ESCs Derived from Skin Cells by Somatic Cell Nuclear Transfer

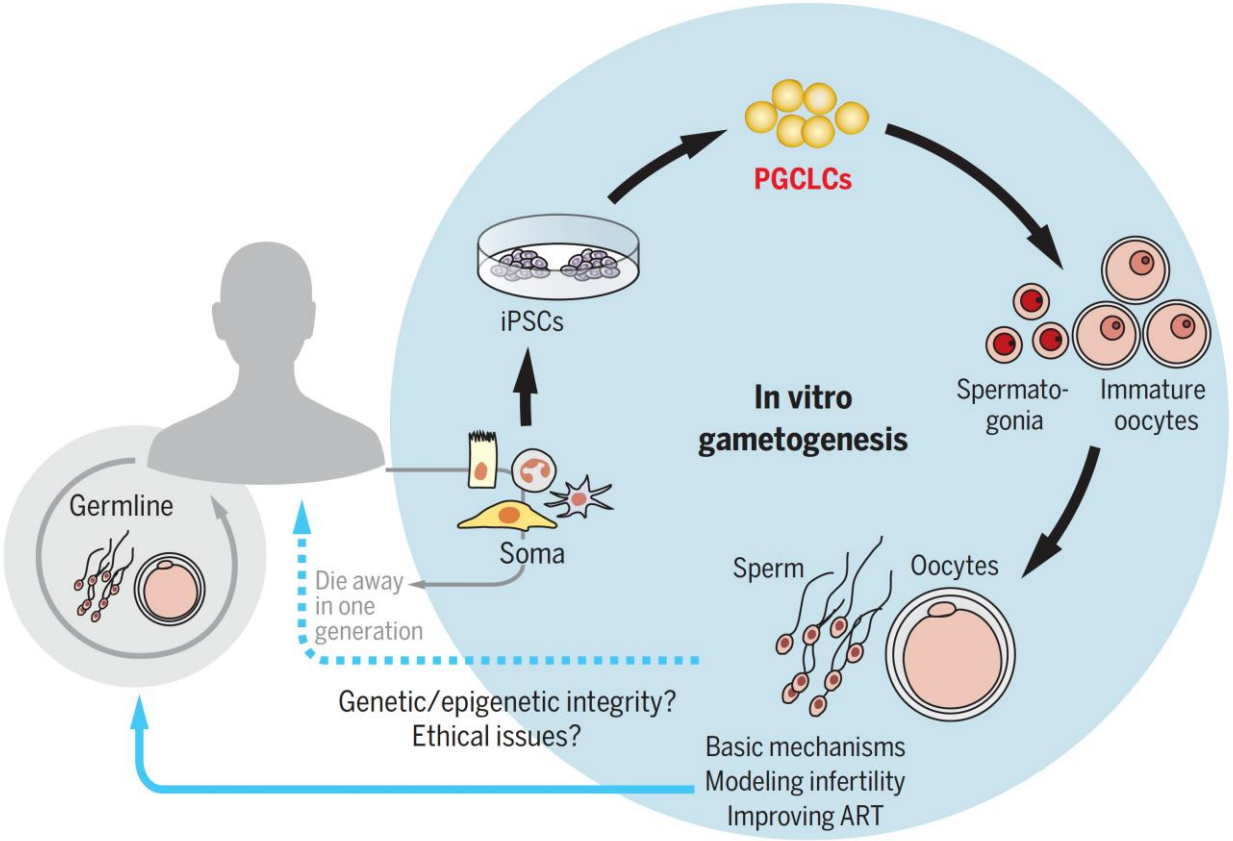


Tachibana et al., Cell, 2013

Reprogramming of Skin Cells to Induced Pluripotent Stem Cells (iPSCs)



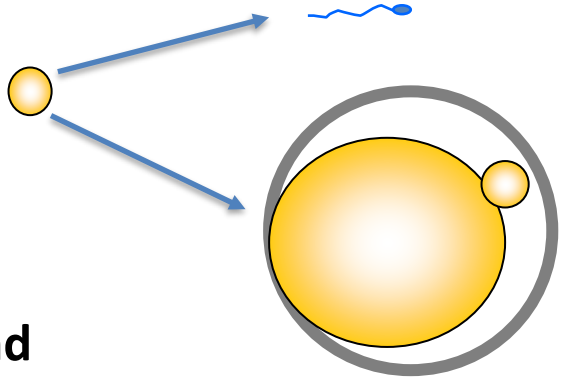
In Vitro Gametogenesis (IVG)



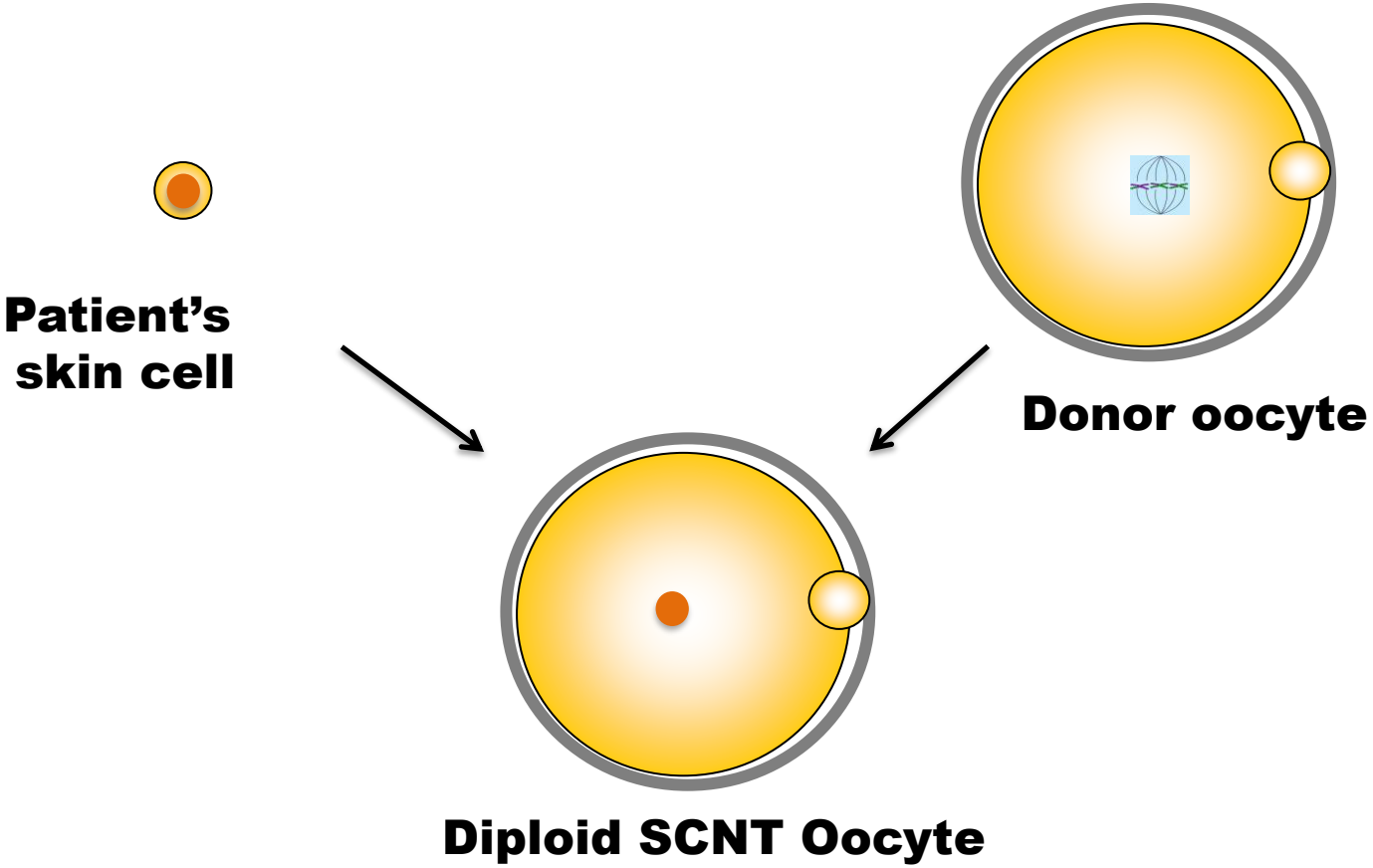
Saitou & Hayashi *Science* 2021

Requirements for somatic cells to become functional gametes

- ✓ Acquiring epigenetic identity of mature sperm or oocytes
- ✓ Haploidy
- ✓ Recombination
- ✓ Imprinting reset
- ✓ Oocytes must acquire cytoplasmic maternal factors and germline mitochondria and mtDNA critical for induction of totipotency and preimplantation embryo development



IVG/SCNT - making oocytes from somatic cells by Somatic Cell Nuclear Transfer

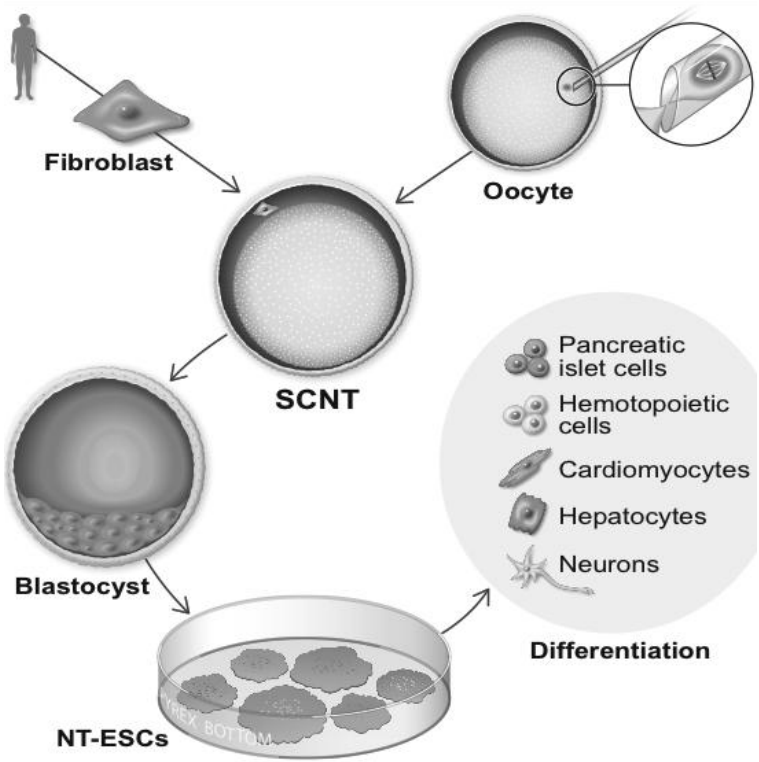


Please cite this article in press as: Tachibana et al., Human Embryonic Stem Cells Derived by Somatic Cell Nuclear Transfer, Cell (2013), http://dx.doi.org/10.1016/j.cell.2013.05.006

Cell

Human Embryonic Stem Cells Derived by Somatic Cell Nuclear Transfer

Masahito Tachibana,[1] Paula Amato,[2] Michelle Sparman,[1] Nuria Marti Gutierrez,[1] Rebecca Tippner-Hedges,[1] Hong Ma,[1] Eunju Kang,[1] Alimujiang Fulati,[1] Hyo-Sang Lee,[1,6] Hathaitip Sritanaudomchai,[3] Keith Masterson,[2] Janine Larson,[2] Deborah Eaton,[2] Karen Sadler-Fredd,[2] David Battaglia,[2] David Lee,[2] Diana Wu,[2] Jeffrey Jensen,[1,4] Phillip Patton,[2] Sumita Gokhale,[5] Richard L. Stouffer,[1,2] Don Wolf,[1] and Shoukhrat Mitalipov[1,2,*]

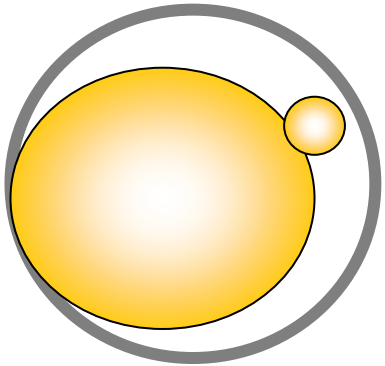


Tachibana et al., Cell, 2013
Ma et. al., Nature, 2014 and 2015
Kang et al., Cell Stem Cell, 2016

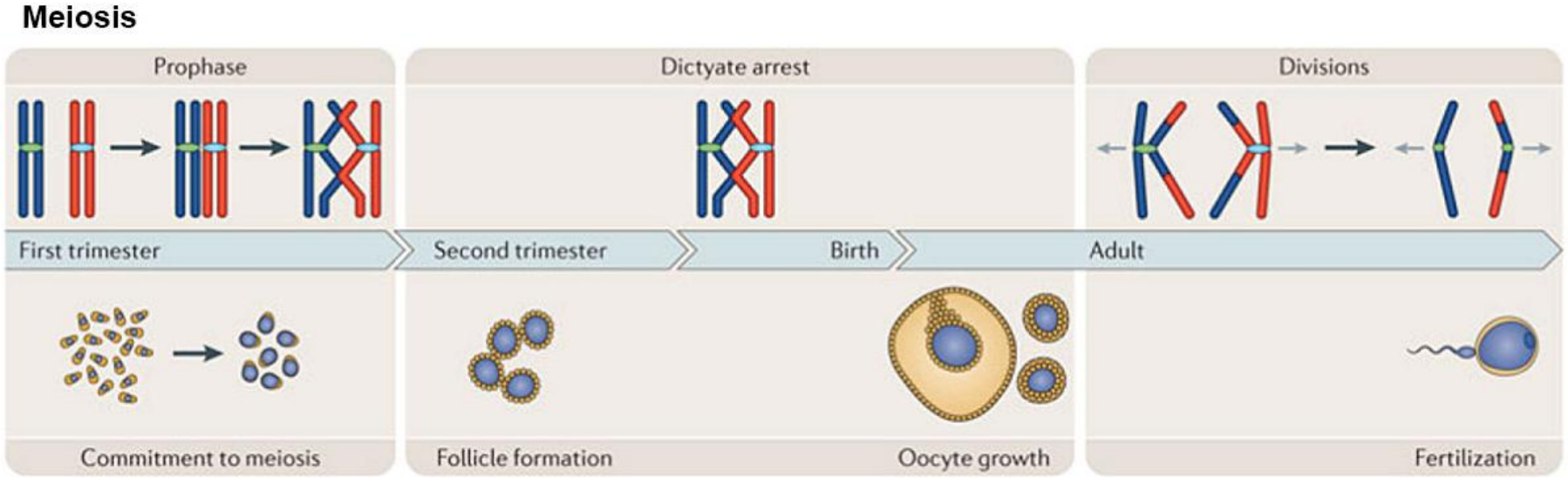
Normal development of offspring derived from diploid SCNT oocytes

Advantages of SCNT-based IVG

- ✓ Epigenetic identity of mature oocytes
- ✓ Cytoplasmic maternal factors of oocytes critical for induction of totipotency and preimplantation embryo development
- ❑ Haploidy
- ❑ Recombination
- ❑ Imprinting reset



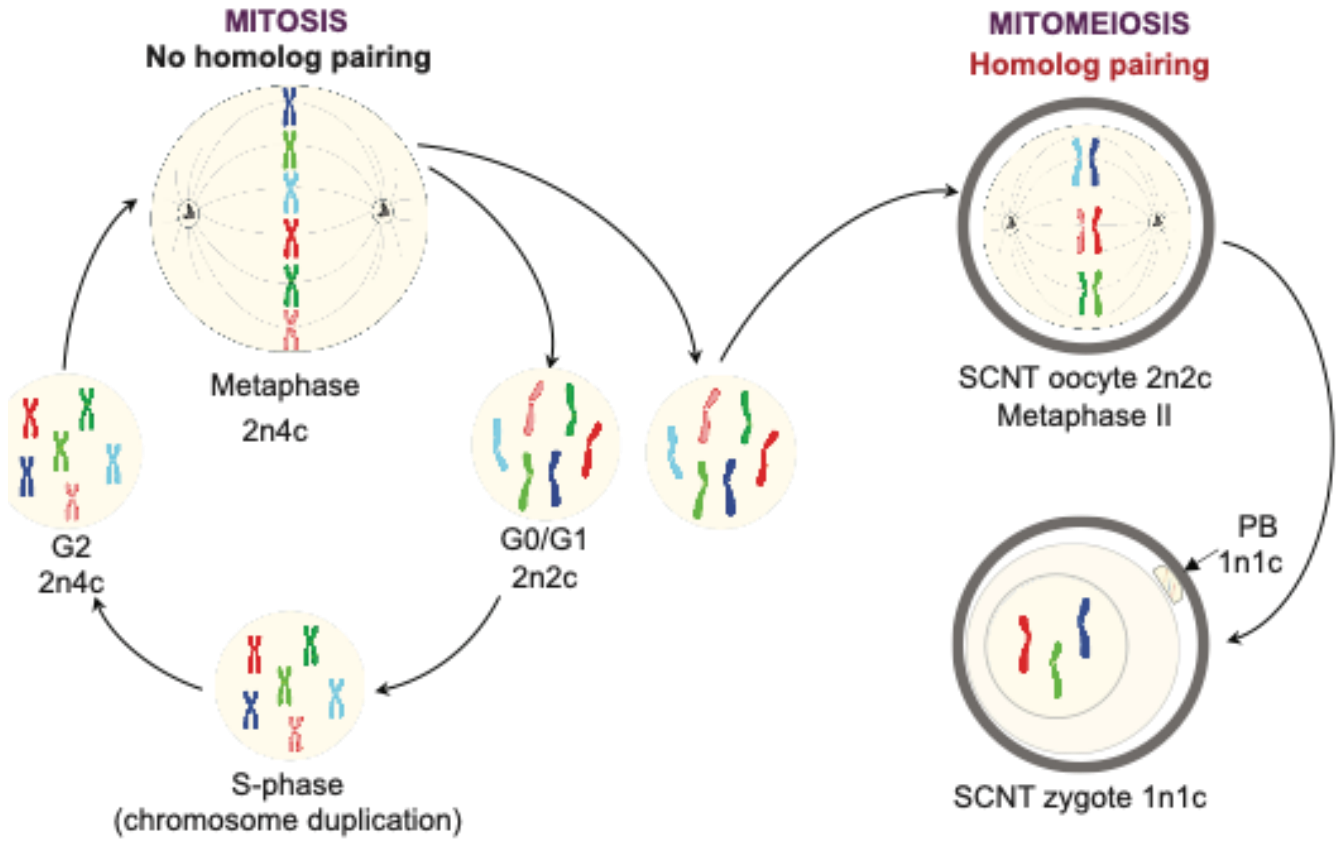
Induction of Haploidy and Crossover Recombination During Female Meiosis



PMID: 22705668
Nature Reviews | Genetics

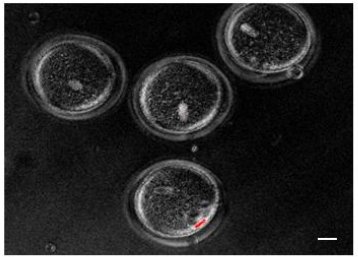
Haploidy induced by experimental reductive cell division

mitomeiosis

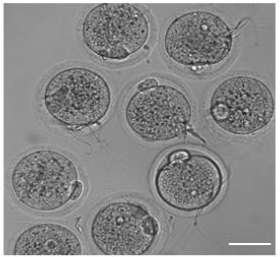


Mikhalchenko et. al., 2024

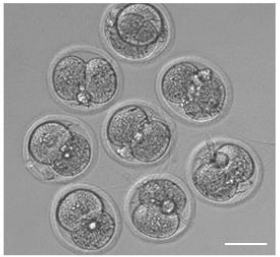
Mouse offspring produced by fertilization of haploid SCNT oocytes



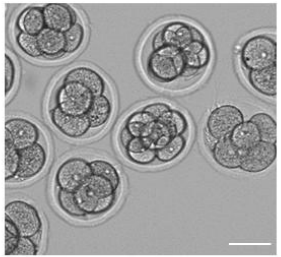
Reconstructed oocytes



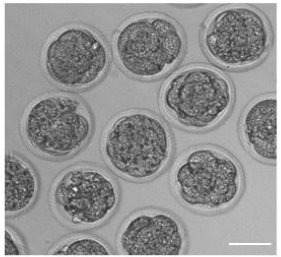
Zygotes



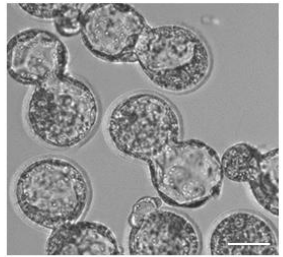
2-cells



4-8-cells



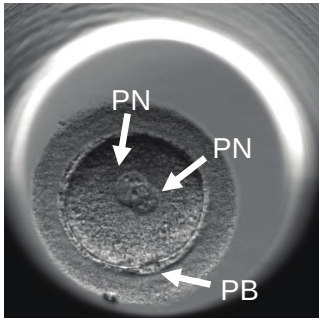
Morula

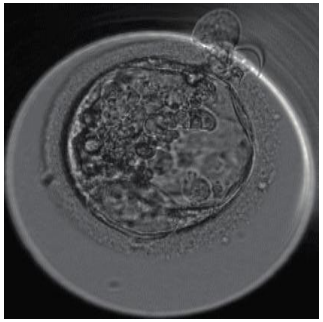


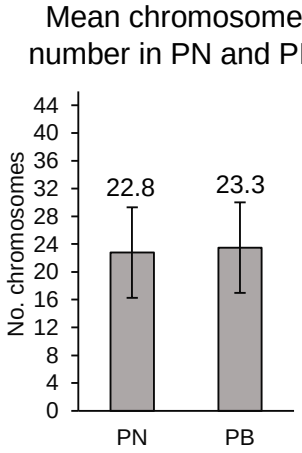
Blastocysts

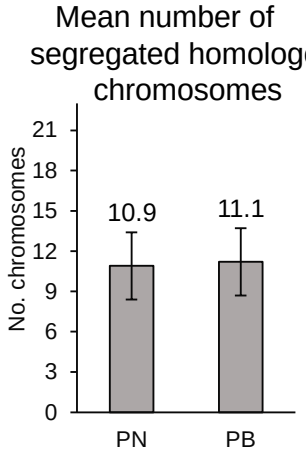
Yeonmi Lee et al., 2022

Fertilization and development of human haploid SCNT oocytes









unpublished

Human IVG by SCNT - Challenges

- ✓ **Improve homologous chromosome recognizing, pairing and synapsis**
- ✓ **Induce crossover recombinations**
- ✓ **Reset imprinting**

OHSU Center for Embryonic Cell and Gene Therapy

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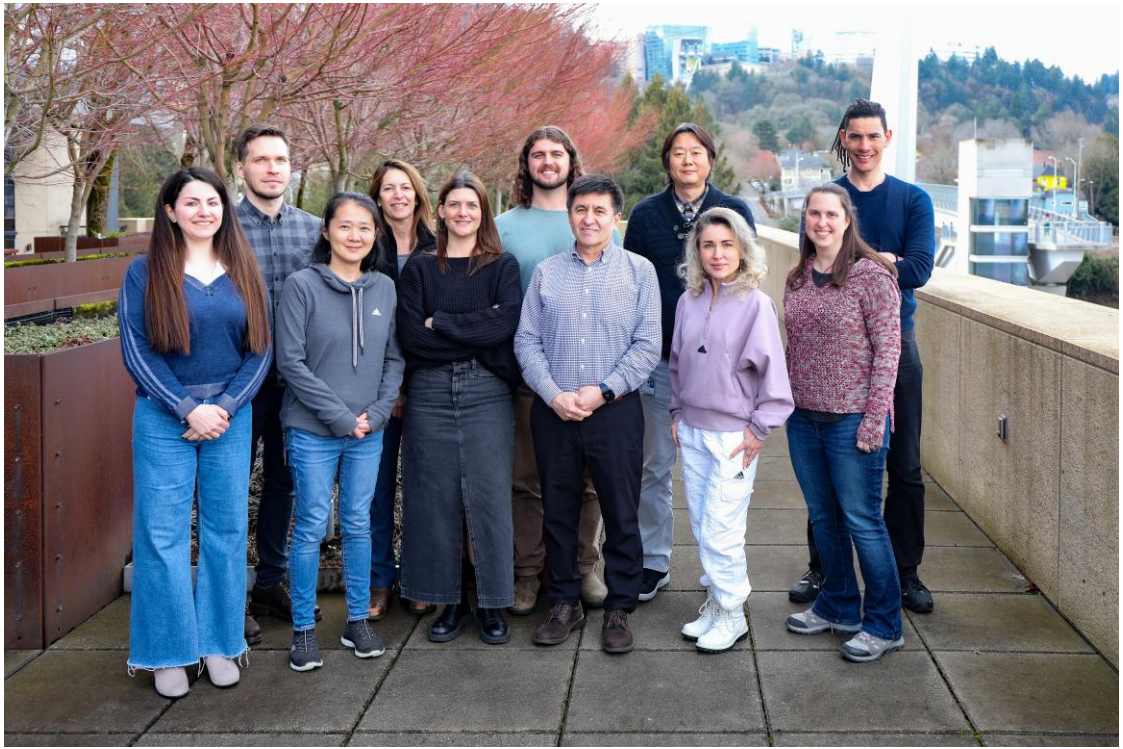

Embryologist and Postdoctoral positions are available