6. Doppelstunde ESF II 2018

Teil 2 Herstellung von Lebewesen aus einzelnen diploiden Zellen (5. bis 7. Doppelstunde)

1. Der weibliche Reproduktionszyklus ex vivo
2. Der männliche Reproduktionszyklus ex vivo
3. Herstellung von Zygoten

} Herstellung von Keimzellen und Zygoten gefolgt von ***in vivo*** Embryogenese!

3.1. Herstellen von bimaternalen (parthenogenetischen) und biandrogenetischen Zygoten und Mäusen (Vorgeschichte: Klonen, parthenog. & androg. Haploide ESCs)

1. Herstellung von Blastozysten aus Stammzellen
2. Herstellung von Plazenten aus Stammzellen
3. Autonome Morphogenese

} ***In vitro*** Embryogenese

4. Ethische und juridische Überlegungen zur Herstellung von Leben

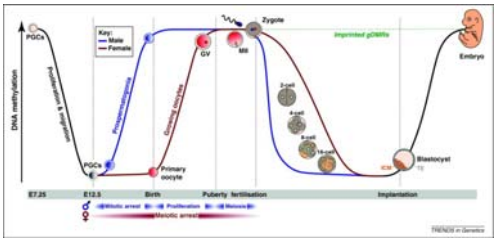
In vitro Reproduktion von Säugetieren inklusive Homo sapiens

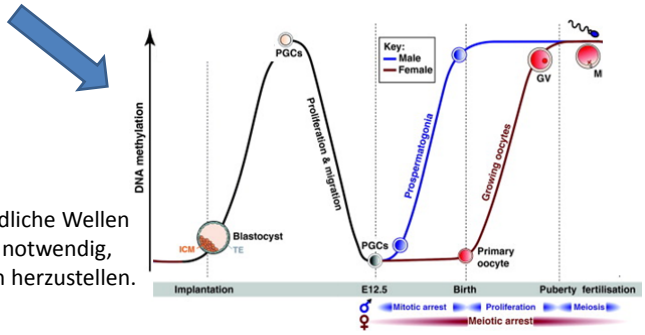
Somatische Zelle → Keimbahnstammzellen (Primordial Germ Cells (PGCs) →

→ Oozyten und Spermien → in vitro Fertilisation (iVF) → künstlicher Uterus

und Plazenta → Embryonal und Fötalentwicklung → Neugeborenes Lebewesen.

Was ist das Problem bei der Ausreifung der Keimzellen?





Es sind zwei unterschiedliche Wellen von DNA Methylierung notwendig, um aus ESCs Keimzellen herzustellen.

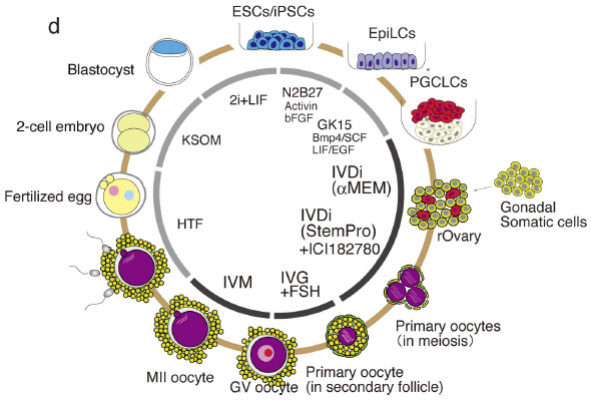
2. Der weibliche Reproduktionszyklus ex vivo

LETTER

doi:10.1038/nature20104

Reconstitution *in vitro* of the entire cycle of the mouse female germ line

Orie Hikabe[1]*, Nobuhiko Hamazaki[1], Go Nagamatsu[1], Yayoi Obata[2], Yuji Hirao[3], Norio Hamada[3,4], So Shimamoto[1], Takuya Imamura[1], Kinichi Nakashima[1], Mitinori Saitou[2,5,6,7,8] & Katsuhiko Hayashi[1,9]*



Are human oocytes from stem cells next?
Johan E J Smitz & Robert B Gilchrist
Nature Biotechnology **volume 34**, pages 1247–1248 (2016)

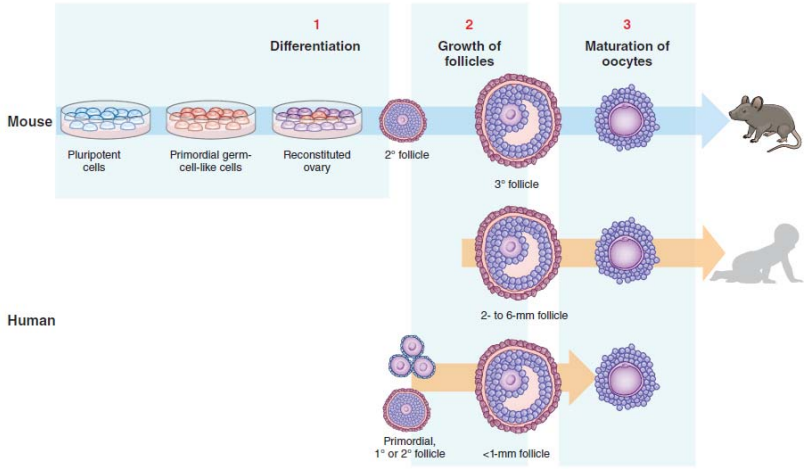
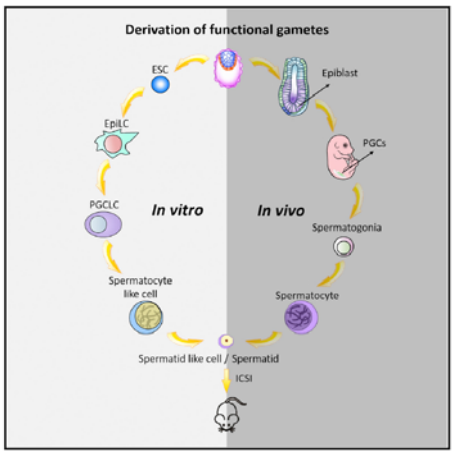


Figure 1 Hikabe *et al.*[1] succeeded in producing mouse oocytes entirely *in vitro* from stem cells. Making human oocytes *in vitro* is likely to be particularly challenging. To date, human pregnancies have been achieved after *in vitro* culture of nearly full-grown oocytes from 2- to 6-mm follicles, but progress in using earlier-stage oocytes has been limited.

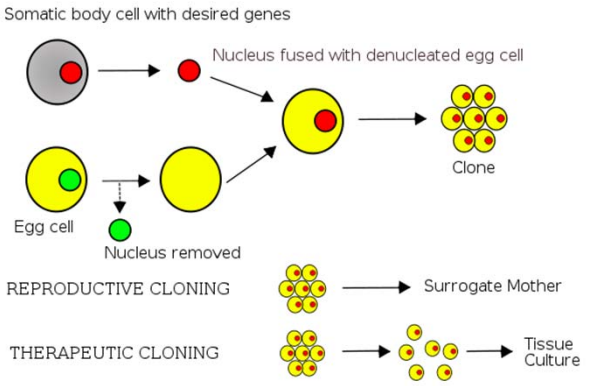
2. Der männliche Reproduktionszyklus ex vivo

Complete Meiosis from Embryonic Stem Cell-Derived Germ Cells In Vitro. Zhou et al., 2016
Cell Stem Cell. 2016 Mar 3;18(3):330-40. doi: 10.1016/j.stem.2016.01.017. Epub 2016 Feb 25.



Link to paper

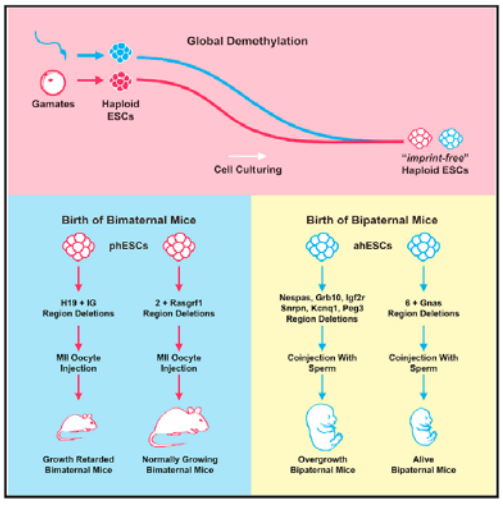
3. Klonen (SCNT)



Wikipedia

Bei Interesse, siehe auch ESFI Vorlesung 4 Dst. 2014

3.1. Bi- maternal und Bi- parternale Mäuse 2018



Generation of Bimaternal and Bipaternal Mice from Hypomethylated Haploid ESCs with Imprinting Region Deletions

Zhi-Kun Li [5]Le-Yun Wang [5]Li-Bin Wang [5]Wei LiQi Zhou [6]Bao-Yang Hu Show all authors Show footnotes

Published:October 11, 2018DOI:https://doi.org/10.1016/j.stem.2018.09.004

- **Highlights**
- •Haploid ESCs display PGC-like methylation profiles following *in vitro* cultivation
- •Parthenogenetic and androgenetic haploid ESCs show different demethylation dynamics
- •phESCs carrying 3 deleted imprinted regions support normal growth of bimaternal mice
- •ahESCs carrying 7 deleted imprinted regions produce live full-term bipaternal mice
- **Summary**
- Unisexual reproduction is widespread among lower vertebrates, but not in mammals. Deletion of the H19 imprinted region in immature oocytes produced bimaternal mice with defective growth; however, bipaternal reproduction has not been previously achieved in mammals. We found that cultured parthenogenetic and androgenetic haploid embryonic stem cells (haESCs) display DNA hypomethylation resembling that of primordial germ cells. Through MII oocyte injection or sperm coinjection with hypomethylated haploid ESCs carrying specific imprinted region deletions, we obtained live bimaternal and bipaternal mice. Deletion of 3 imprinted regions in parthenogenetic haploid ESCs restored normal growth of fertile bimaternal mice, whereas deletion of 7 imprinted regions in androgenetic haploid ESCs enabled production of live bipaternal mice that died shortly after birth. Phenotypic analyses of organ and body size of these mice support the genetic conflict theory of genomic imprinting. Taken together, our results highlight the factors necessary for crossing same-sex reproduction barriers in mammals.