Stem Cell Biology and Technology

Prof. David Suter

Gene regulation and cell identity

BIO-447 2024/2025

Other teachers

Prof. David Suter
Gene regulation & Cell identity

Introduction, plasticity; tools; neural stem cells

Prof. Wouter Karthaus
Endocrine therapy resistance and molecular genetics

Adult stem cells; niche of adult stem cells; stem cell bioengineering

Prof. Freddy Radtke

ES, iPS, cloning and nuclear reprogramming; Intestinal stem cells; Cancer stem cells

Prof. Olaia Naveiras
CHUV, UNIL

Clinical impact of Stem cells

Dr. Jean-Francois Mayol
Flow cytometry Facility UNIL

Flow cytometry and FACS

SCHEDULE

Stem Cell Course Winter Semester 2024/25

Dates	Subject	Teacher
13.09.	Introduction to stem cells	David Suter
20.09.	Plasticity	David Suter
27.09.	Neural stem cells	David Suter
04.10.	Tools in stem cell biology	David Suter
11.10.	Adult stem cells	Wouter Karthaus
18.10.	Flow Cytometry and FACS	Jean-Francois Mayol
01.11.	Niches of adult stem cells	Wouter Karthaus
08.11.	Stem cell bioengineering (Mimicking the niches)	Wouter Karthaus
15.11.	ES, iPS, cloning and nuclear reprogramming	Freddy Radtke
22.11.	Intestinal stem cells	Freddy Radtke
29.11.	Clinical Impact of stem cells I	Olaia Naveiras
06.12.	Clinical Impact of stem cells II	Shukry James Habib
13.12	Cancer stem cells	Freddy Radtke
22.12.	free	free

Enrollment key: IPS&Co

Exercises and exam

Exercise time

- **No exercises**
- **But teachers might ask you to read something**

Exam

- **Multiple choice**
- **Might be some short open answer questions**

Additional resources

- **International Society for Stem Cell Research (ISSCR):** http://www.isscr.org/
  - tons of interesting background information (Glossary, answers to FAQs, movies, etc.)

- **Nature REPORTS stem cells:** https://www.nature.com/stemcells/index.html
  - always updated on latest key papers in the field
  - interviews with leading researchers in the field

- **NIH Stem Cell Information:** http://stemcells.nih.gov/

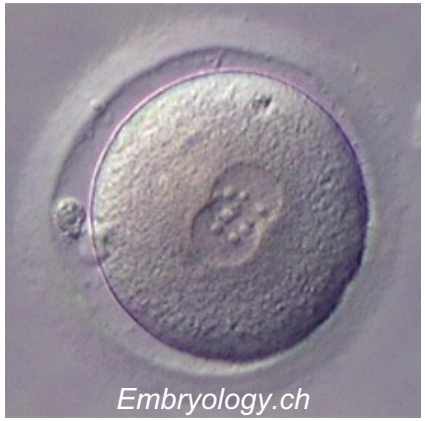
- **EuroStemCell:** http://www.eurostemcell.org/

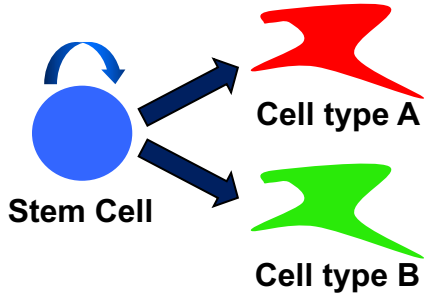
- **StemBook:** https://www.stembook.org/

Stem cell definition

One genome, from one to hundreds cell types

1 genome, 1 cell



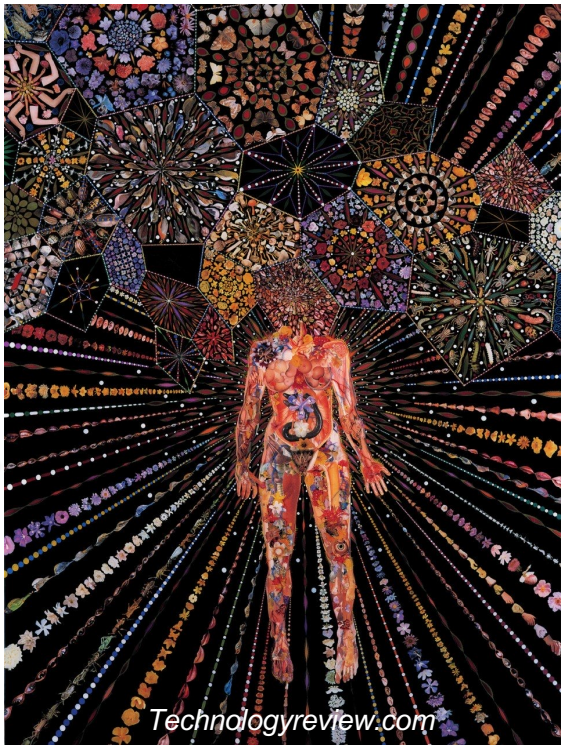


?

Developmental disorders

Cancer

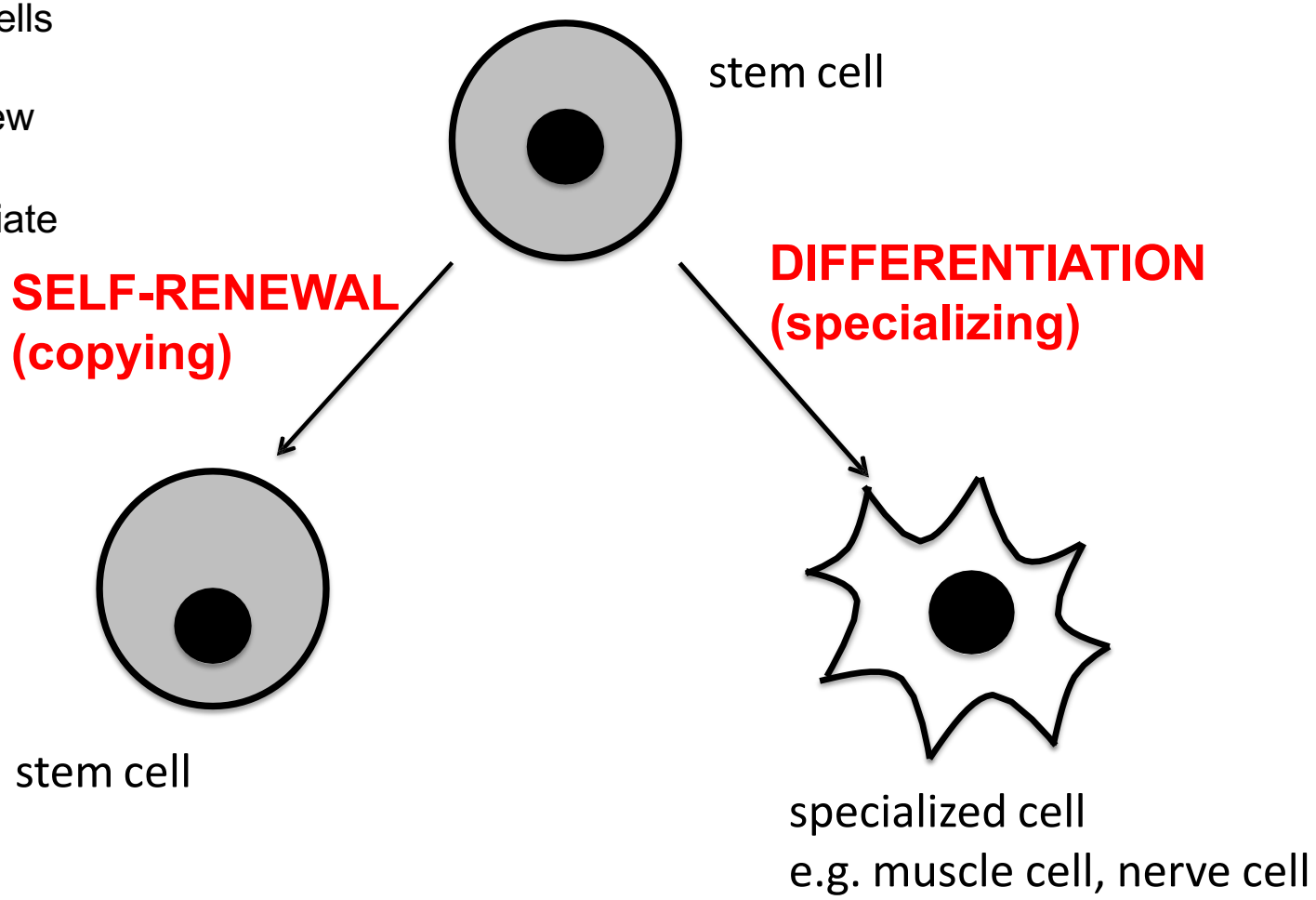
1 genome, >200 cell types



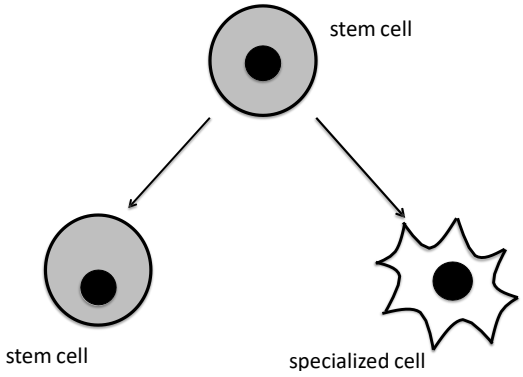
What is a stem cell?

Stem cells are:

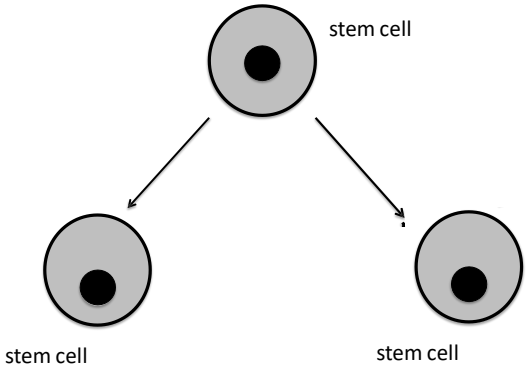
- Unspecialized cells
- Able to self-renew
- Able to differentiate



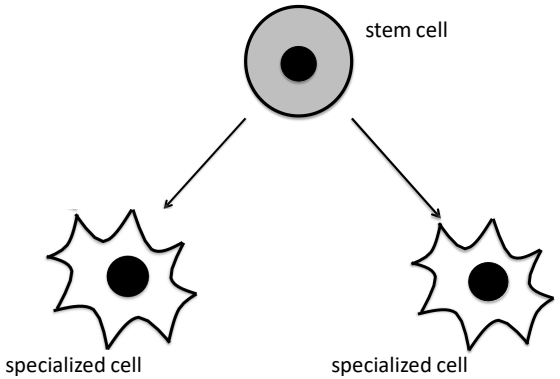
Different types of stem cell divisions



Asymmetric division

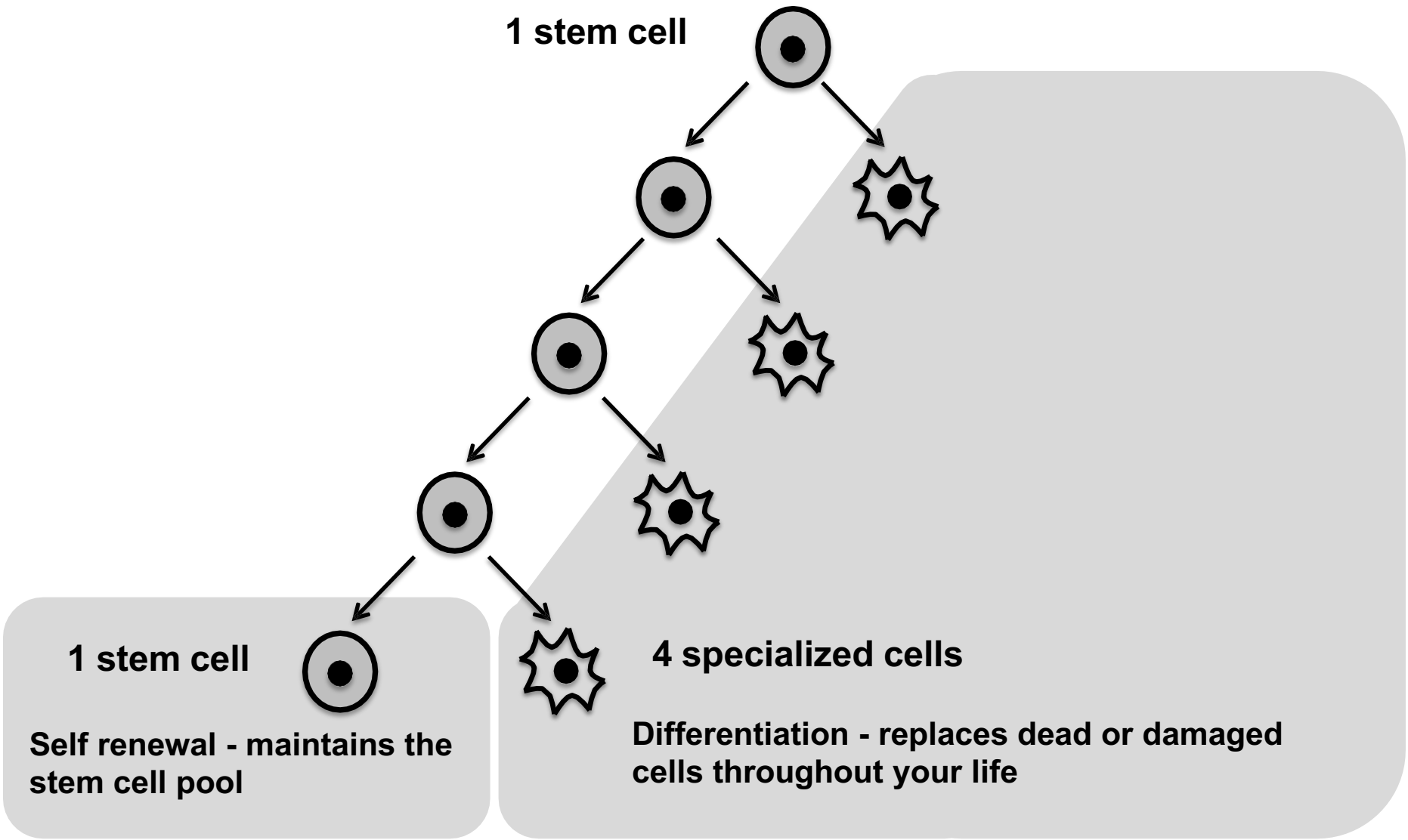


Symmetric self-renewal division



Symmetric differentiation division

Why self-renewal and differentiation?



History of stem cell research

Stem cell research was born in the aftermath of Hiroshima and Nagasaki (6. and 9. August 1945)

1961-63: Till & McCulloch → “colony forming cells” in the bone marrow self-renew and differentiate

1962: J.D.Gurdon → cell specialisation is reversible (2012 Nobel prize)

1981: Evans & Kaufman → first embryonic stem cells (ESCs) isolated from mouse blastocysts (2007 Nobel prize)

1989: Smithies & Capecchi → first knock-out mice (2007 Nobel prize)

1996: Wilmut → first mammal cloned from adult cells

1998: Thomson → derivation of first human ESC line

2006: Takahashi & Yamanaka → generation of induced pluripotent stem (iPS) cells (2012 Nobel prize)

A bit of early history:
Ramalho-Santos & Willenbring 2007: https://www.cell.com/cell-stem-cell/fulltext/S1934-5909(07)00019-7
Maehle 2011: https://royalsocietypublishing.org/doi/10.1098/rsnr.2011.0023

The experiments of Till and McCulloch 1961-1963

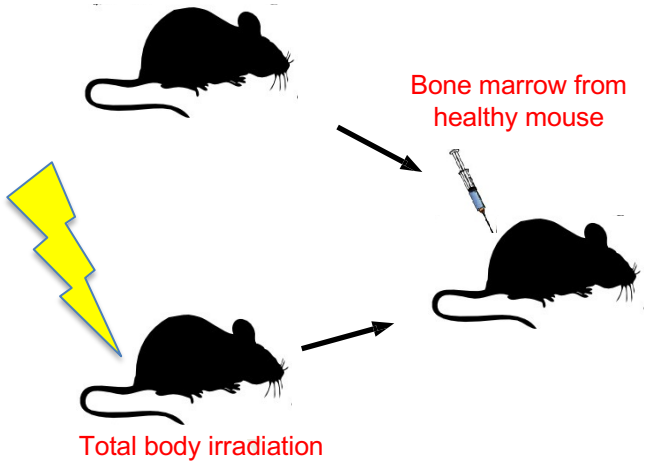
RADIATION RESEARCH **14,** 213–222 (1961)

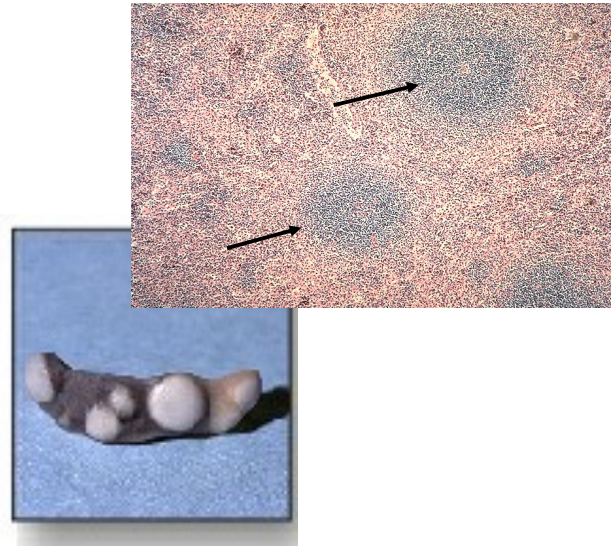
A Direct Measurement of the Radiation Sensitivity of Normal Mouse Bone Marrow Cells[1]

J. E. TILL AND E. A. McCULLOCH

Department of Medical Biophysics, University of Toronto, and the Divisions of Biological Research and Physics of the Ontario Cancer Institute, Toronto, Ontario

Cells with **differentiation** capacity, "colony forming cells" are present in the bone marrow





Myeloid, erythroid, lymphoid precursors

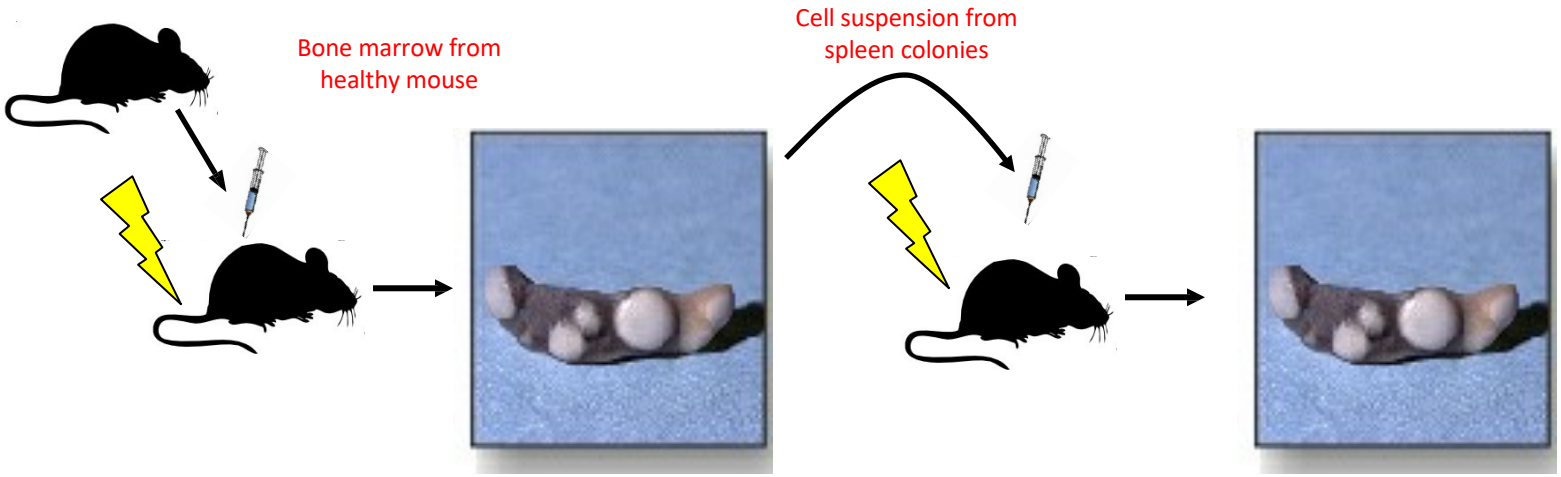
The experiments of Till and McCulloch 1961-1963

The Distribution of Colony-forming Cells Among Spleen Colonies

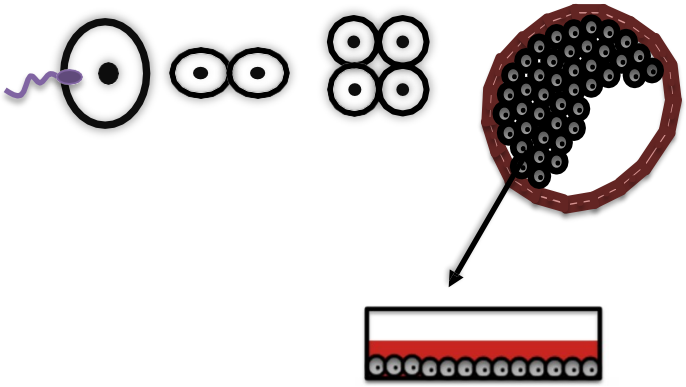
L. SIMINOVITCH, E. A. McCULLOCH AND J. E. TILL
Department of Medical Biophysics, University of Toronto, and The Ontario Cancer Institute, Toronto, Ontario

JOURNAL OF CELLULAR AND COMPARATIVE PHYSIOLOGY
Vol. 62, No. 3, December 1963

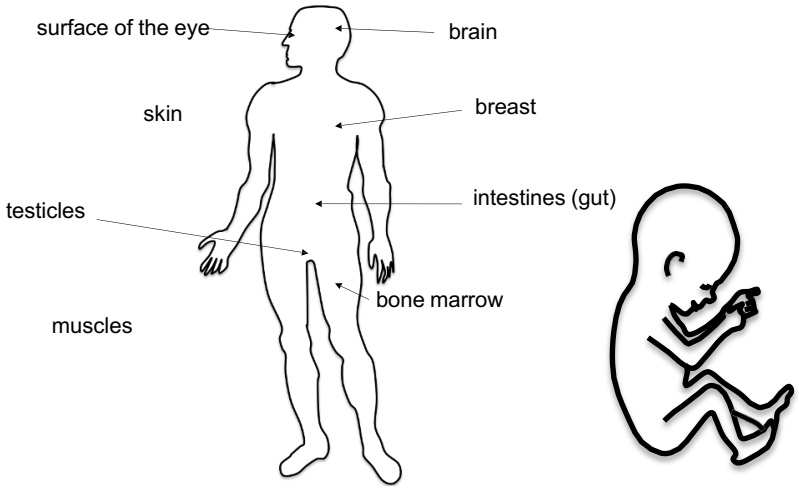
- Perform serial transplantation
- The “colony-forming cells” can renew themselves and form new “colony-forming cells”
- First assay to test **self-renewal** capacity



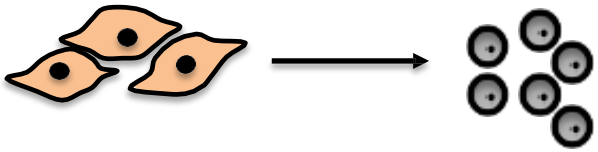
Different types of stem cells exist



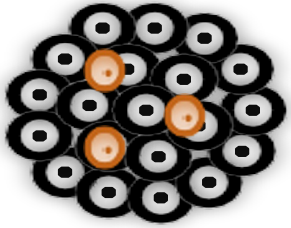
Embryonic stem cells



Tissue stem cells



Induced pluripotent stem cells



Cancer stem cells

Stem cell definition - Potency

Potency	**A measure of how many types of specialized cells a stem cell can make**
Totipotent	**Can make all types of cells in the body and placenta** **Note: The zygote is totipotent, but it is not a stem cell!**
Pluripotent	**Can make all types of cells in the body, but not placenta** **Embryonic stem cells are pluripotent**
Multipotent	**Can make multiple types of specialized cells, but not all types** **Tissue stem cells are multipotent**
Unipotent	**Can make a single type of specialized cells** **Spermatogonial stem cells are unipotent**

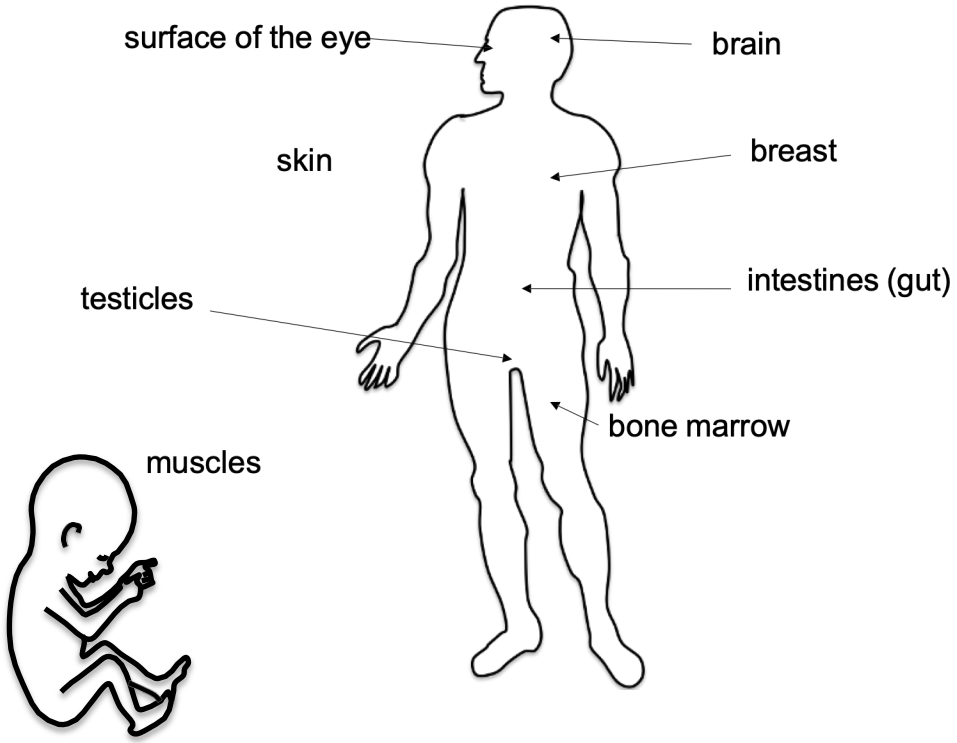
Types of stem cells

1. Tissue stem cells

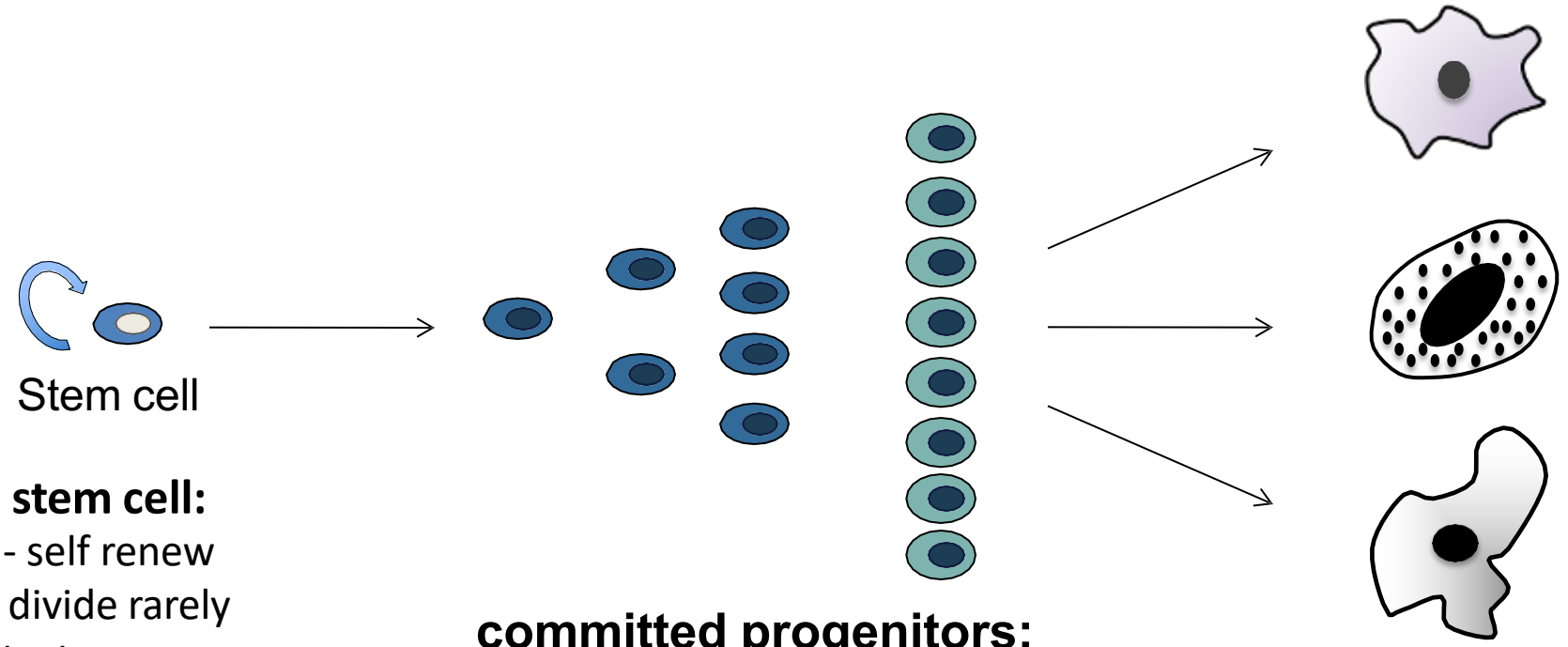
Tissue stem cells - characteristics

- Found in adult and fetal tissues
- **Multipotent** (can make multiple types of specialized cells, but not all types)
- Usually rare
- Rarely divide

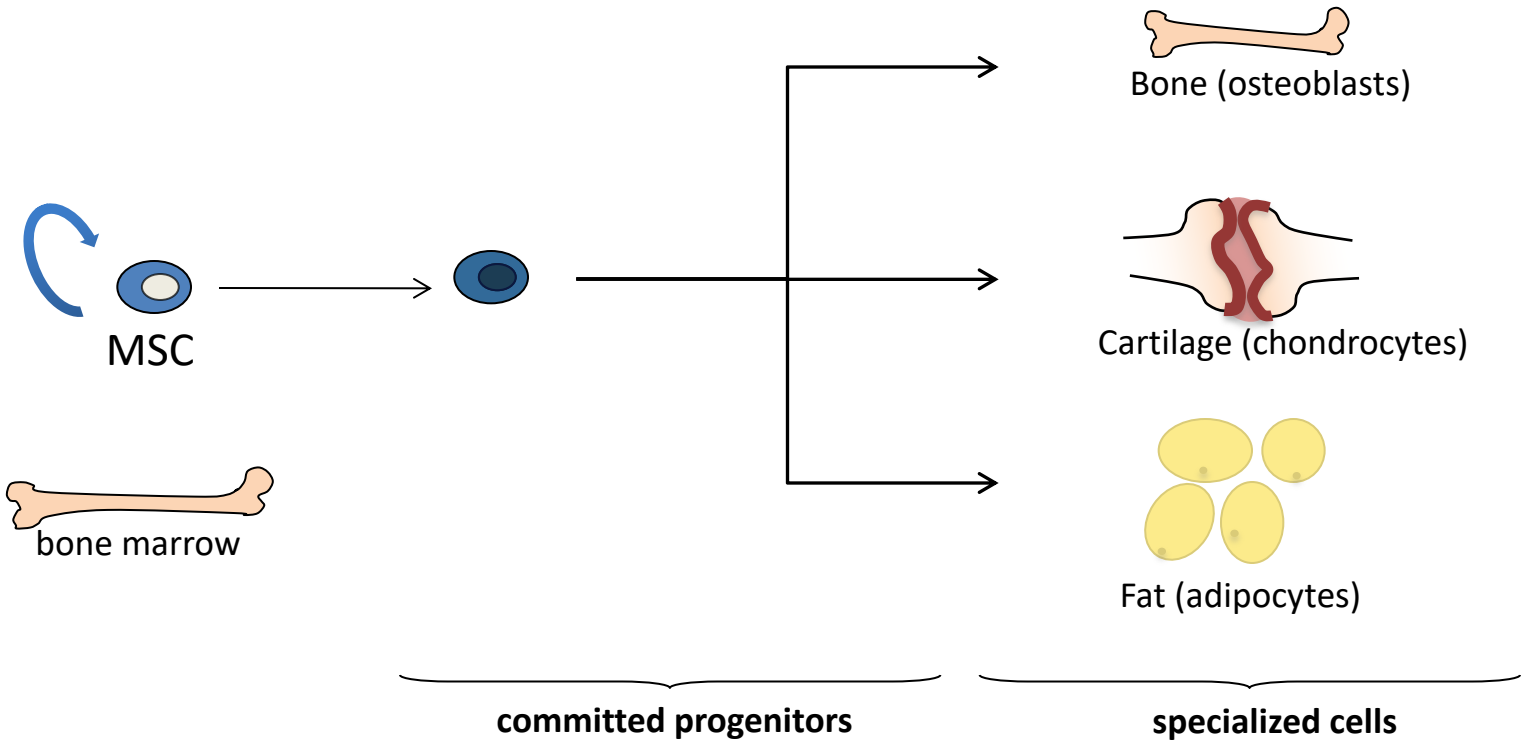
→ how can tissues still be renewed?



Principles of renewing tissues - stem cell hierarchies

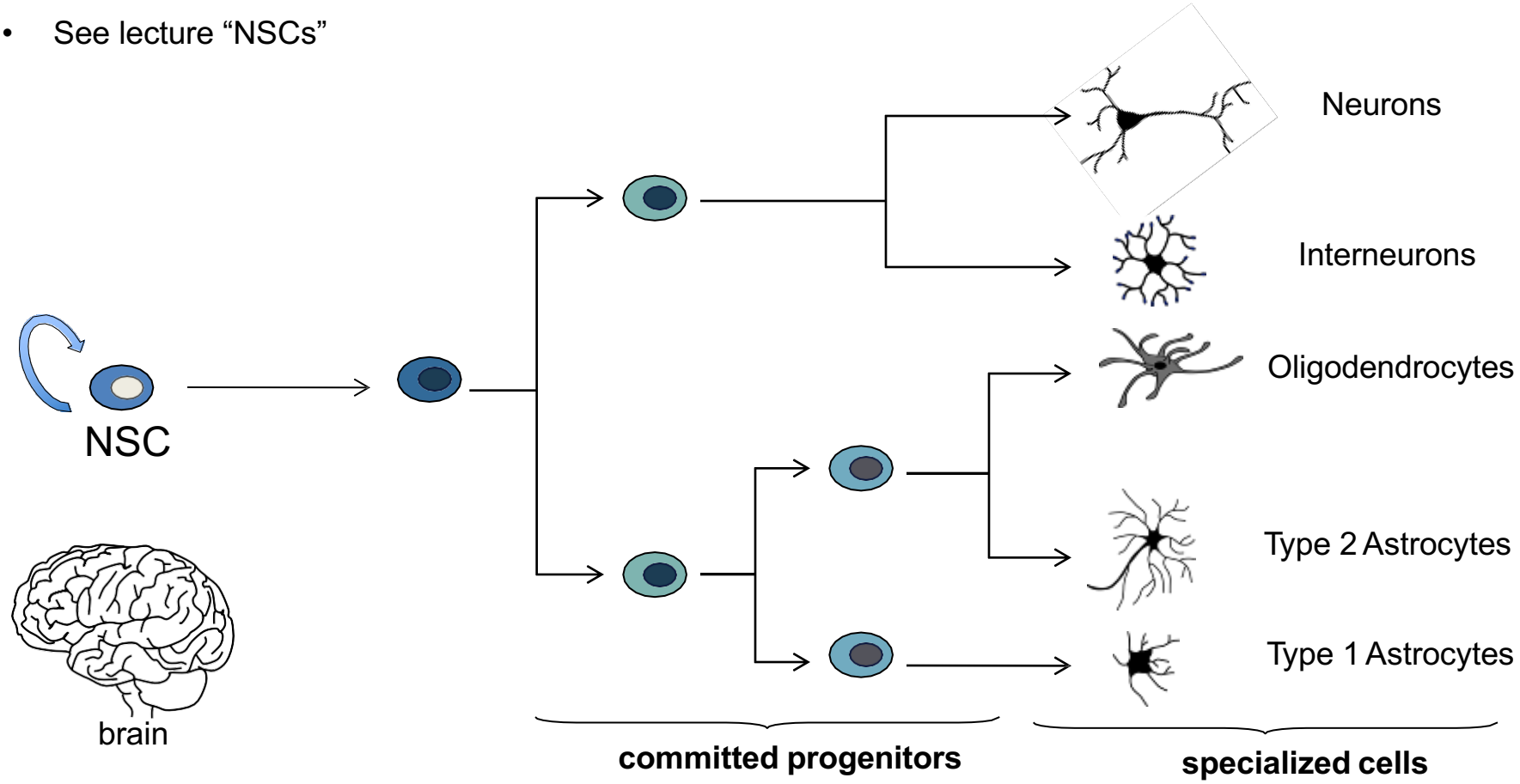


Mesenchymal stem cells (MSCs)



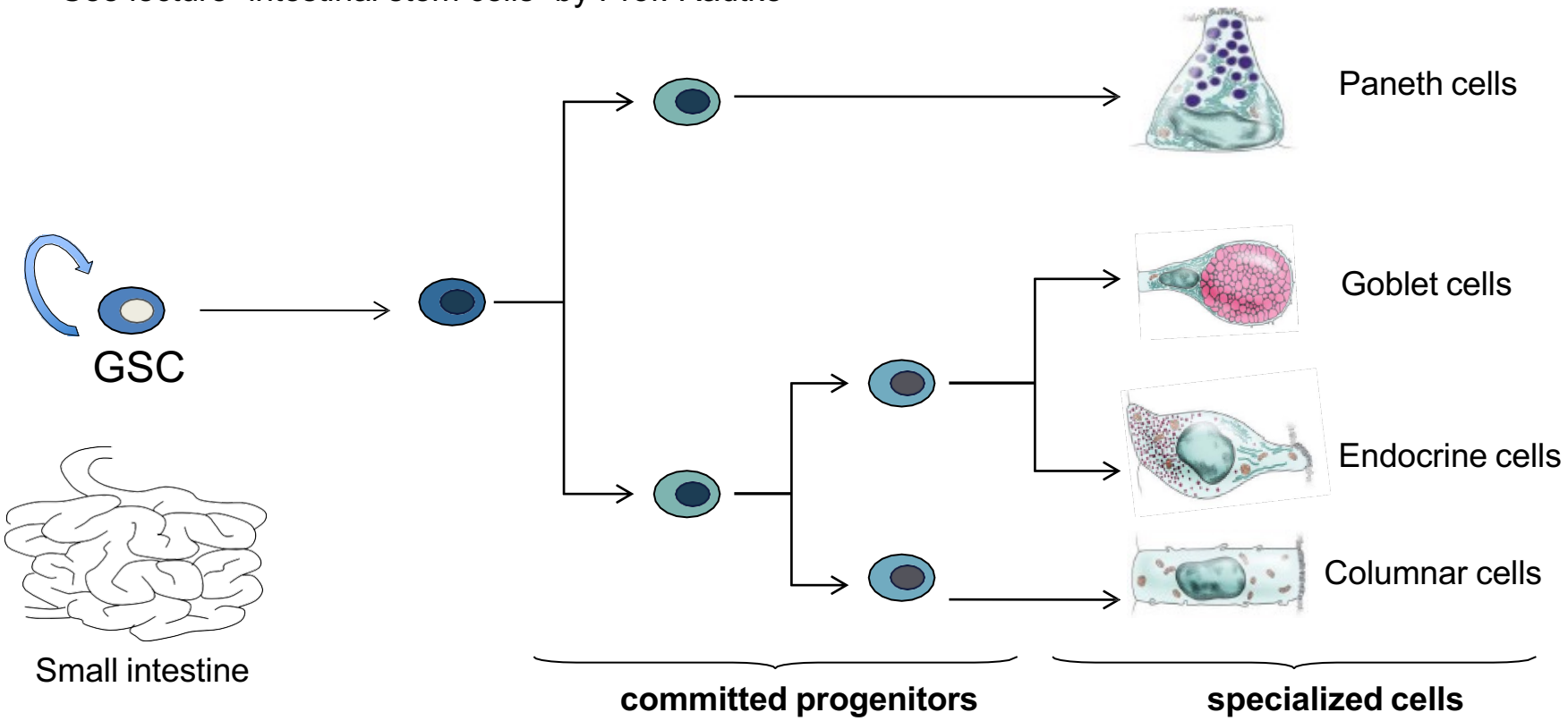
Neural stem cells (NSCs)

- See lecture “NSCs”



Gut or intestinal stem cells (GSCs)

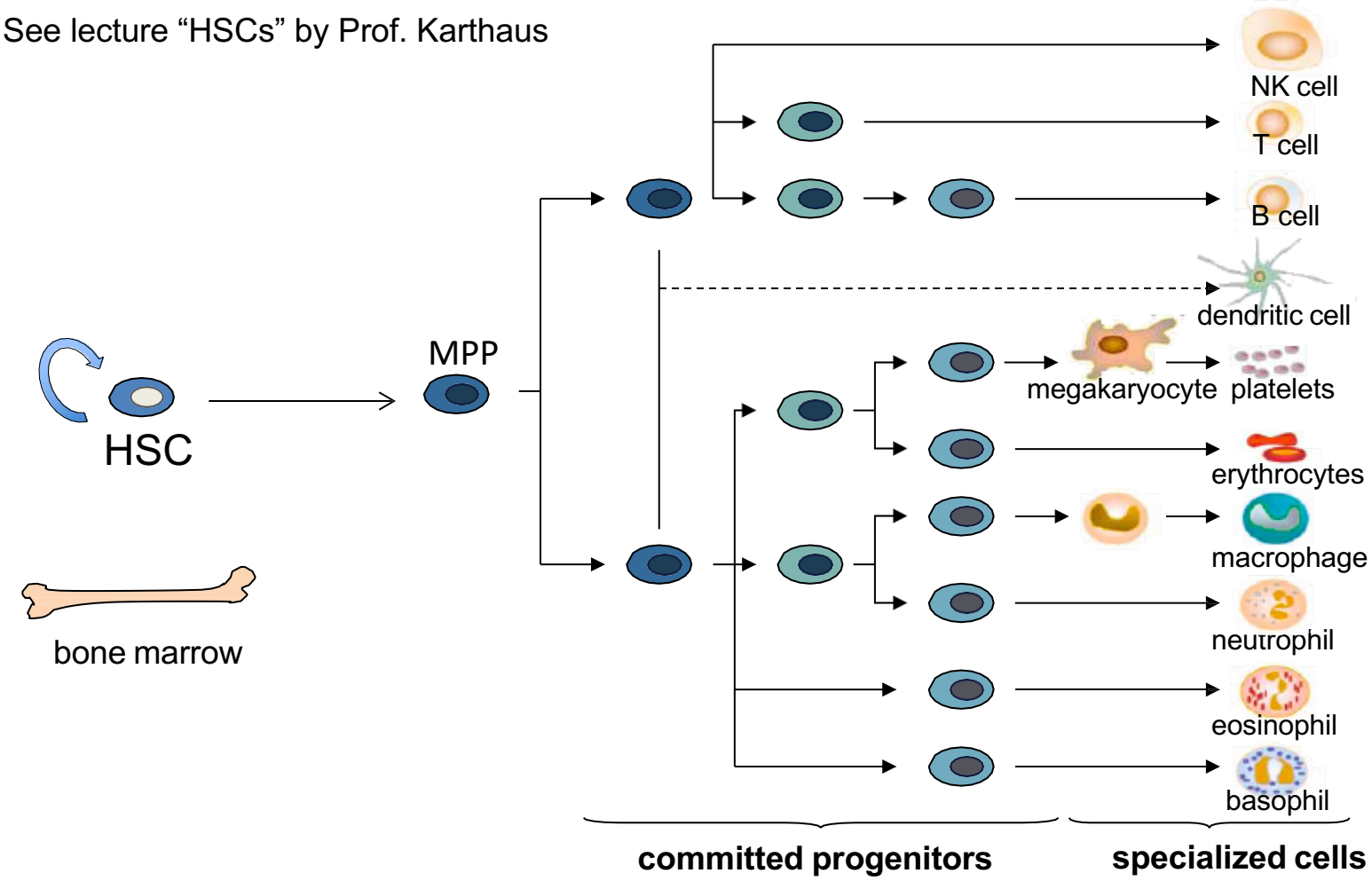
- See lecture “intestinal stem cells” by Prof. Radtke



https://www.youtube.com/watch?v=qq5k1sWqLO0

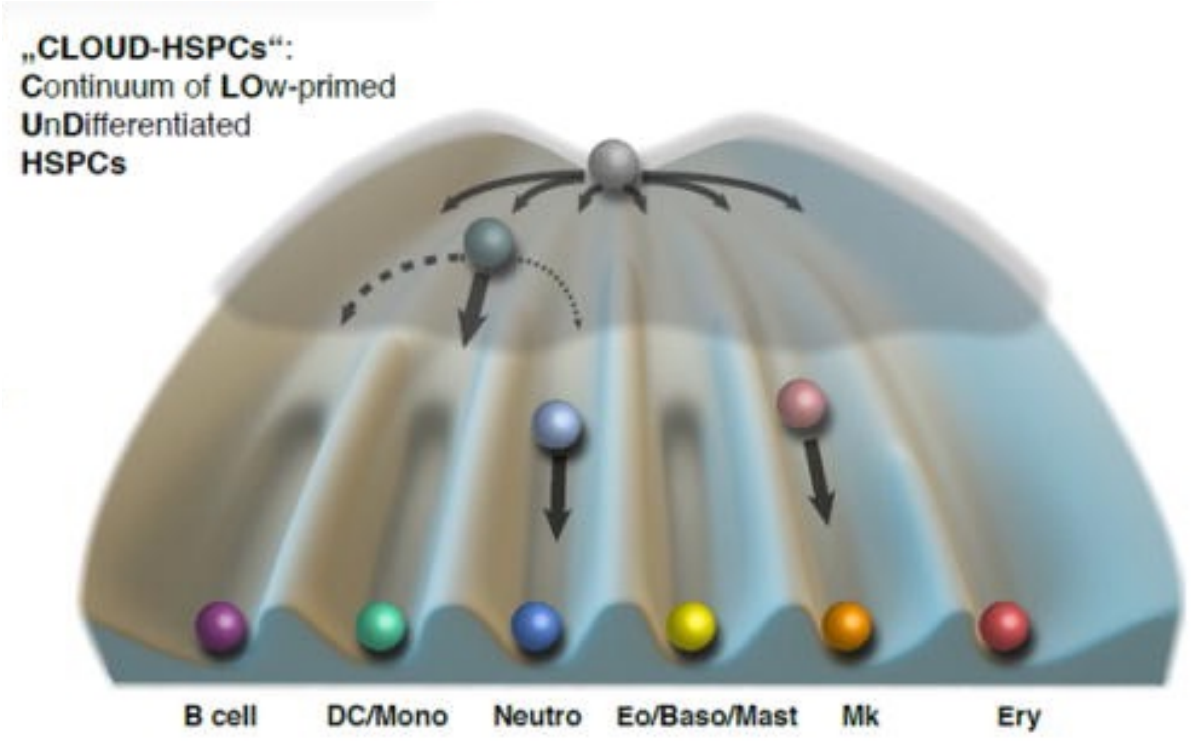
Hematopoietic stem cells (HSCs)

- See lecture “HSCs” by Prof. Karthaus



Tissue stem cell hierarchies are debated

ScRNAseq and other single cell techniques reveal a continuum of committed progenitors



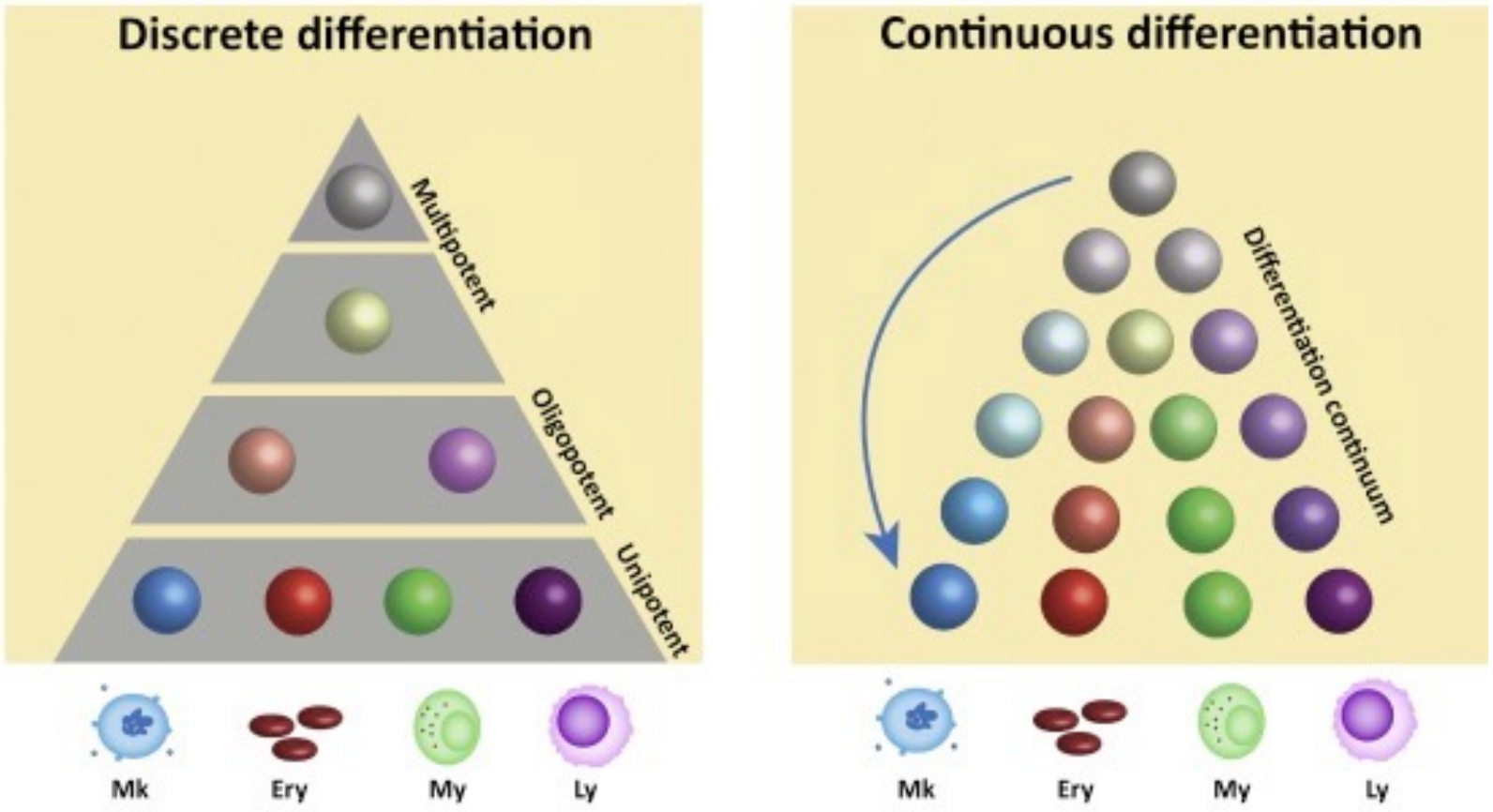
› Nat Cell Biol. 2017 Apr;19(4):271-281. doi: 10.1038/ncb3493. Epub 2017 Mar 20.

Human haematopoietic stem cell lineage commitment is a continuous process

Lars Velten [1], Simon F Haas [2 3 4], Simon Raffel [2 4 5], Sandra Blaszkiewicz [2 3], Saiful Islam [6], Bianca P Hennig [1], Christoph Hirche [2 3], Christoph Lutz [5], Eike C Buss [5], Daniel Nowak [7], Tobias Boch [7], Wolf-Karsten Hofmann [7], Anthony D Ho [5], Wolfgang Huber [1], Andreas Trumpp [2 4 8], Marieke A G Essers [2 3], Lars M Steinmetz [1 6 9]

Distinct lineages emerge directly from CLOUD-HSPCs without passing through a series of discrete, stable progenitors

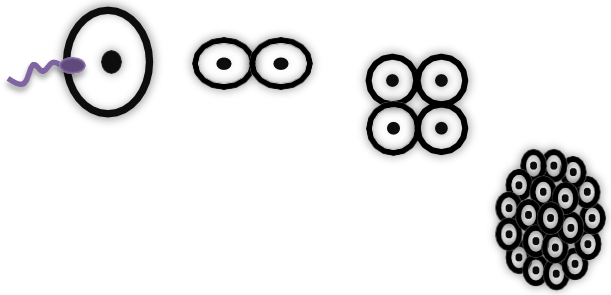
Tissue stem cell hierarchies are debated

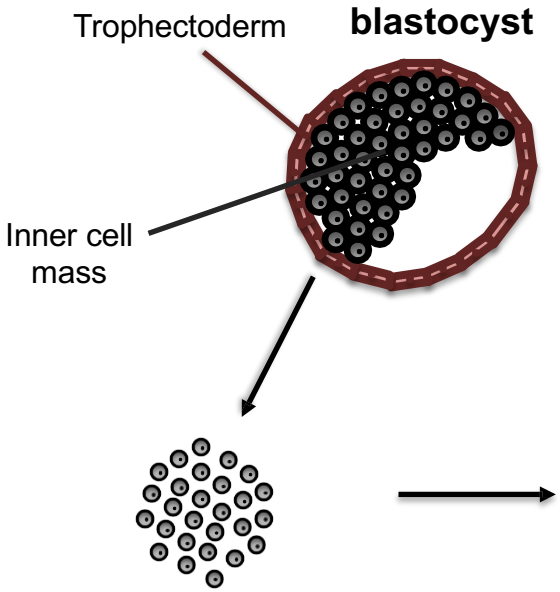


Zhang et al., Trends in Cell Bio 2018

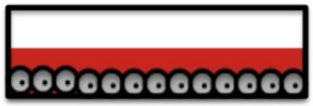
2. Embryonic stem cells (ESCs)

Embryonic stem cells - characteristics



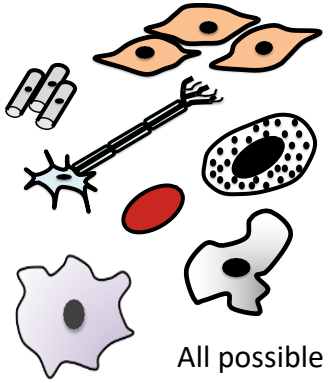


embryonic stem cells taken from the inner cell mass



culture in the lab to grow more cells

differentiation

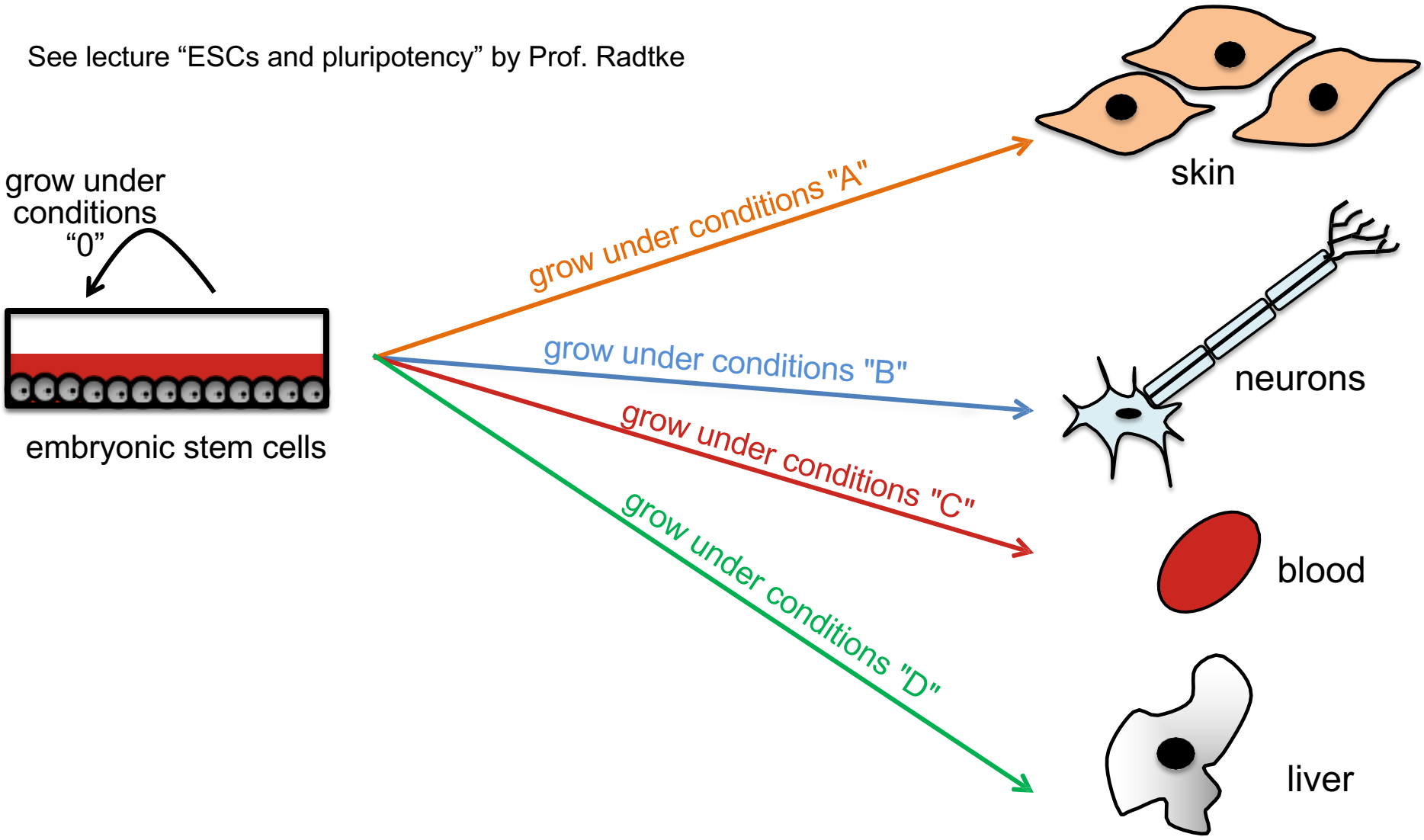


All possible types of specialized cells

- Derived from the inner cell mass of a blastocyst
- **<u>Pluripotent</u>** (can differentiate in all types of cells in the body, but not placenta)
- Ability to self-renew indefinitely
- Divide rapidly
- Can be grown in the lab in a defined medium

Embryonic stem cell differentiation

- See lecture “ESCs and pluripotency” by Prof. Radtke



https://www.youtube.com/watch?v=0iQqOuINeZI

Nobel Prize in Physiology or Medicine 2007

Medicine

The Nobel Prize in Physiology or Medicine 2007

"for their discoveries of principles for introducing specific gene modifications in mice by the use of embryonic stem cells"

Photo: Tim Roberts/PR Newswire, © HHMI

Mario R. Capecchi

1/3 of the prize

USA

University of Utah
Salt Lake City, UT, USA;
Howard Hughes Medical Institute

b. 1937
(in Italy)

Photo: The Press Association Limited

Sir Martin J. Evans

1/3 of the prize

United Kingdom

Cardiff University
Cardiff, United Kingdom

b. 1941

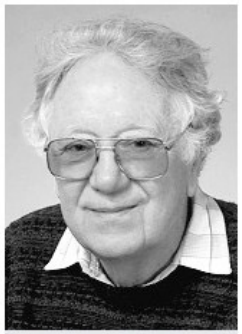


Photo: Scanpix/Dan Sears

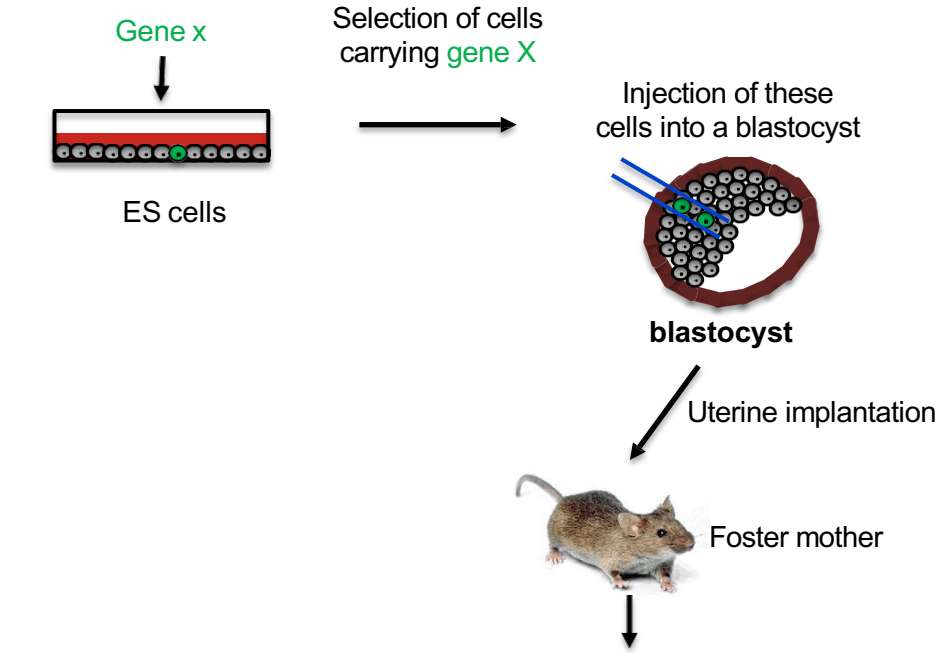
Oliver Smithies

1/3 of the prize

USA

University of North Carolina at Chapel Hill
Chapel Hill, NC, USA

b. 1925
(in United Kingdom)



3. Induced pluripotent stem cells (iPS)

Nuclear reprogramming

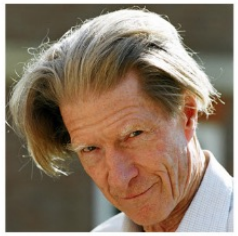
- 1962: Sir John B Gurdon challenged dogma that specialised cells are irreversibly committed to their fate

[J. Embryol. exp. Morph., Vol. 10, Part 4, pp. 622-40 December 1962]

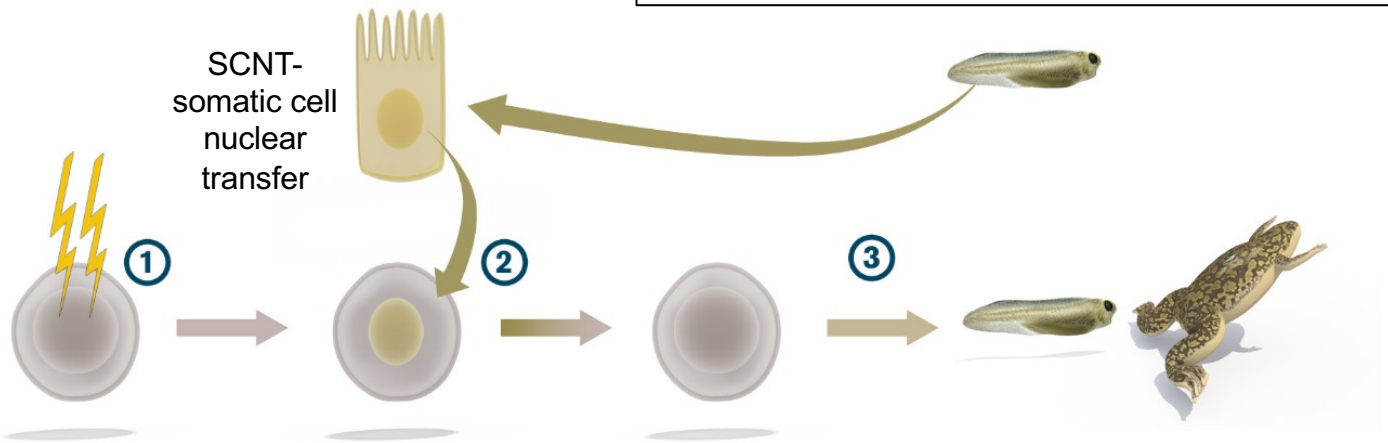
The Developmental Capacity of Nuclei taken from Intestinal Epithelium Cells of Feeding Tadpoles

by J. B. GURDON[1]

From the Embryology Laboratory, Department of Zoology, Oxford



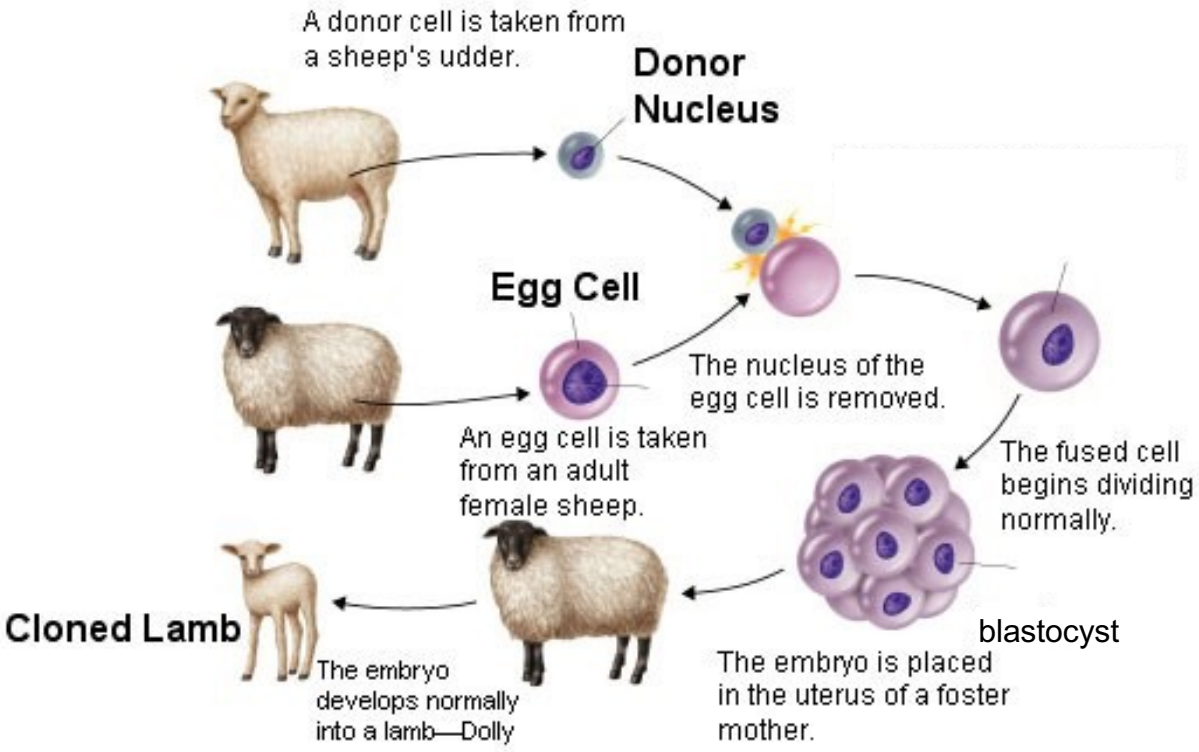
John B. Gurdon



John B. Gurdon eliminated the nucleus of a frog egg cell (1) and replaced it with the nucleus from a specialised cell taken from a tadpole (2). The modified egg developed into a normal tadpole (3). Subsequent nuclear transfer experiments have generated cloned mammals (4).

nobelprize.org/pressrelease

Reproductive cloning: e.g. Dolly the sheep



NATURE | VOL 385 | 27 FEBRUARY 1997

Viable offspring derived from fetal and adult mammalian cells

I. Wilmut, A. E. Schnieke*, J. McWhir, A. J. Kind* & K. H. S. Campbell

Roslin Institute (Edinburgh), Roslin, Midlothian EH25 9PS, UK
** PPL Therapeutics, Roslin, Midlothian EH25 9PP, UK*

July 5, 1996: First mammal cloned from adult cells

Since Dolly: Used in cows, mice, rats, goats, pigs, rabbits, cats, mule, horse and dog, etc.

Reproductive cloning: e.g. Dolly

SINOGENE 希诺谷 ABOUT US PET CLONING CELL PRESERVATION CLIENT STORIES HOW IT WORKS BLOG CONTACT US

SINOGENE PET CLONING SERVICE

Losing a pet is heartbreaking, but SINOGENE provides technological solutions to clone your pets by using a procedure of somatic cell nuclear transfer. As the science of pet cloning has progressed from the past years, SINOGENE provides a platform of gene edition technology, DNA preservation, DNA preservation, and cloning of cat, dog and horse. We are complied with EU regulatory standards and China Animal Welfare Legislation to maintain trust and ensure integrity with customers and society.

Cat Cloning READ MORE

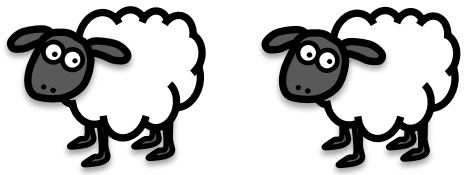
Dog Cloning READ MORE

Horse Cloning READ MORE

Careful!

There are two VERY different types of cloning:

Reproductive cloning

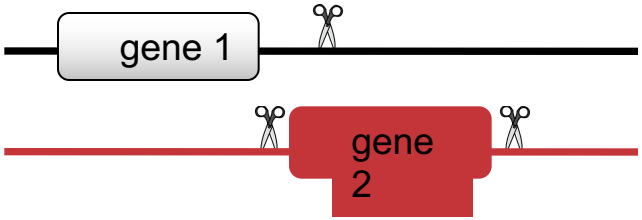


Use to make two identical individuals

Very difficult to do

Illegal to do on humans

Molecular cloning

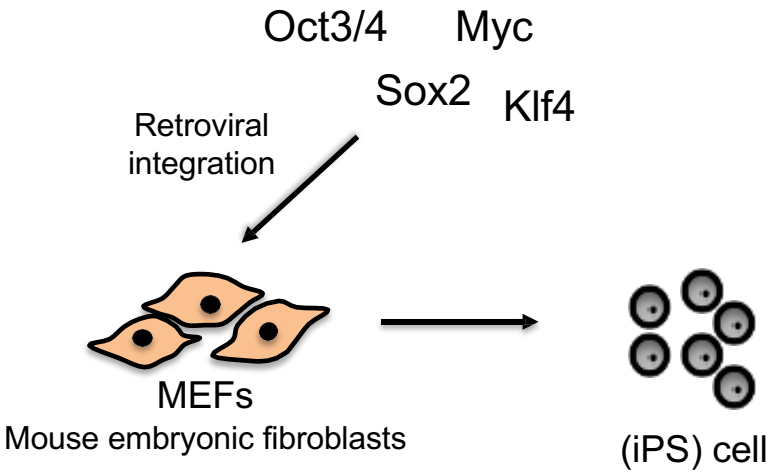


Use to study what a gene does

Routine in biology labs

Induced pluripotent stem cells discovery

- See lecture “nuclear reprogramming and iPS” by Prof. Radtke

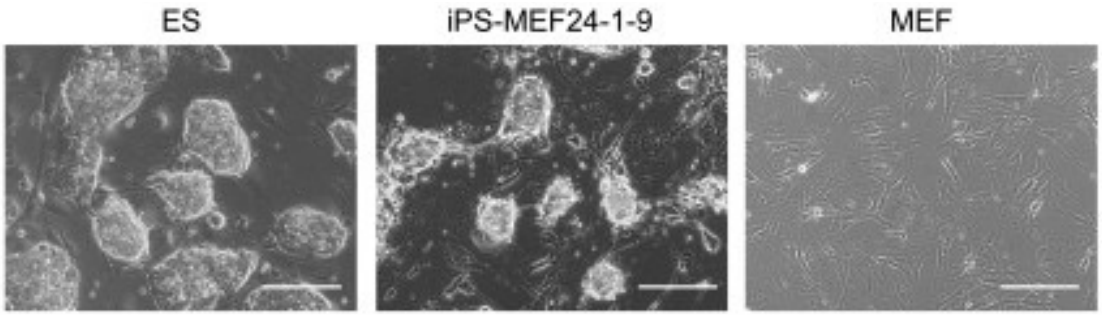


Induction of Pluripotent Stem Cells from Mouse Embryonic and Adult Fibroblast Cultures by Defined Factors

Kazutoshi Takahashi[1] and Shinya Yamanaka[1,2,*]
[1]Department of Stem Cell Biology, Institute for Frontier Medical Sciences, Kyoto University, Kyoto 606-8507, Japan
[2]CREST, Japan Science and Technology Agency, Kawaguchi 332-0012, Japan
*Contact: yamanaka@frontier.kyoto-u.ac.jp
DOI 10.1016/j.cell.2006.07.024

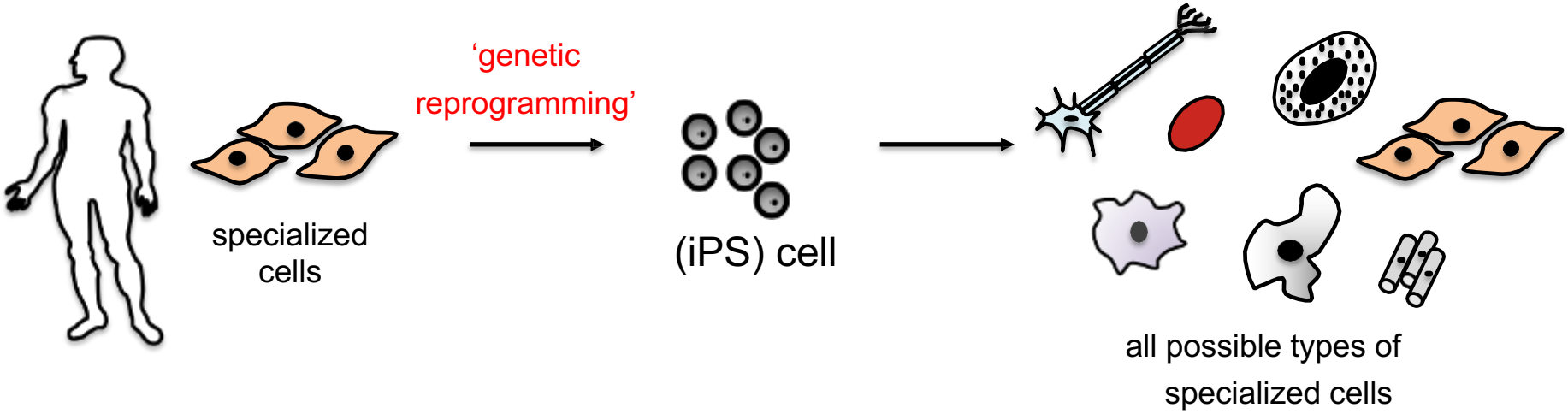
Shinya Yamanaka

- From an original set of 24, 4 factors were sufficient to generate pluripotent cells from MEFs
- Reprogramming still long and inefficient process



https://www.cell.com/action/showPdf?pii=S0092-8674%2806%2900976-7

Induced pluripotent stem cells (iPS) - characteristics



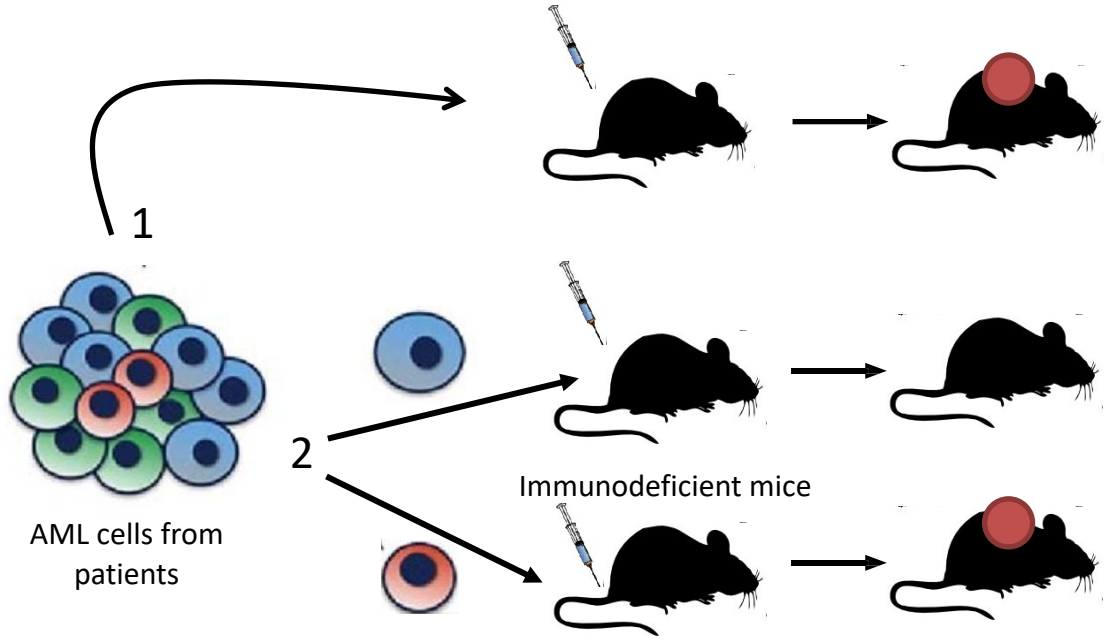
- Behave like ESCs
- **Pluripotent** (can make all types of cells in the body, but not placenta)
- Divide rapidly
- Self-renew indefinitely
- Artificial system
- No embryonic stem cells are needed

To summarize

	Embryonic stem cells (ESCs)	Tissue stem cells	Induced pluripotent stem cells (iPS)
Artificial system	yes	no	yes
Pluripotent	yes	no	yes
Efficient differentiation	no	yes	no
Efficient expansion in culture	yes	no	yes
Rare cell type	no	yes	yes
Immune-compatible	no	no	yes
Teratoma risk	yes	no	yes

4. Cancer stem cells

Cancer stem cell model - key experiment



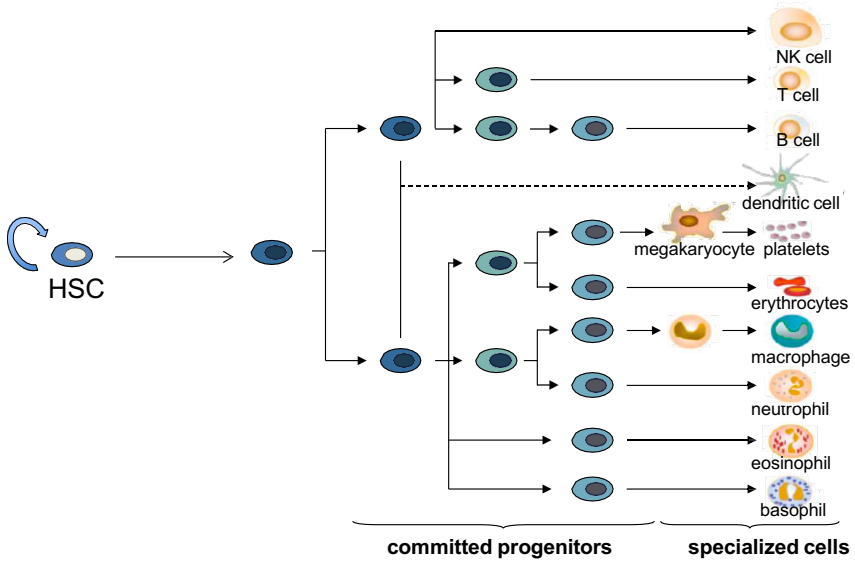
A cell initiating human acute myeloid leukaemia after transplantation into SCID mice

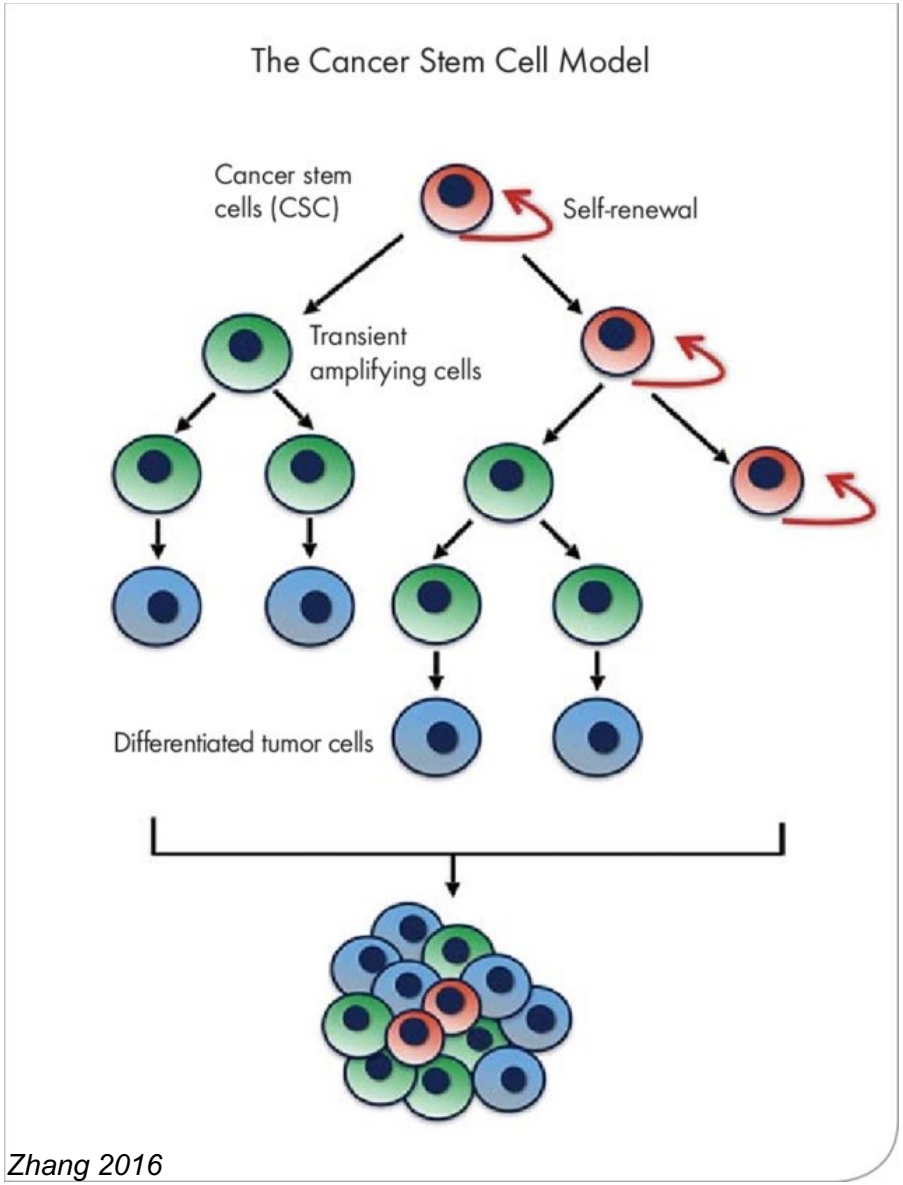
Tsvee Lapidot, Christian Sirard, Josef Vormoor, Barbara Murdoch, Trang Hoang*, Julio Caceres-Cortes*, Mark Minden†, Bruce Paterson‡, Michael A. Caligiuri§ & John E. Dick‖

NATURE VOL 367 · 17 FEBRUARY 1994

- Only a very low fraction of tumour cells can reinitiate the disease
- Cancer stem cells have a high self-renewal capacity
- They share features of tissue stem cells:
  - Rare
  - Rare division
  - Self-renewal
  - Differentiation capacity

Cancer stem cell model explaining tumor heterogeneity



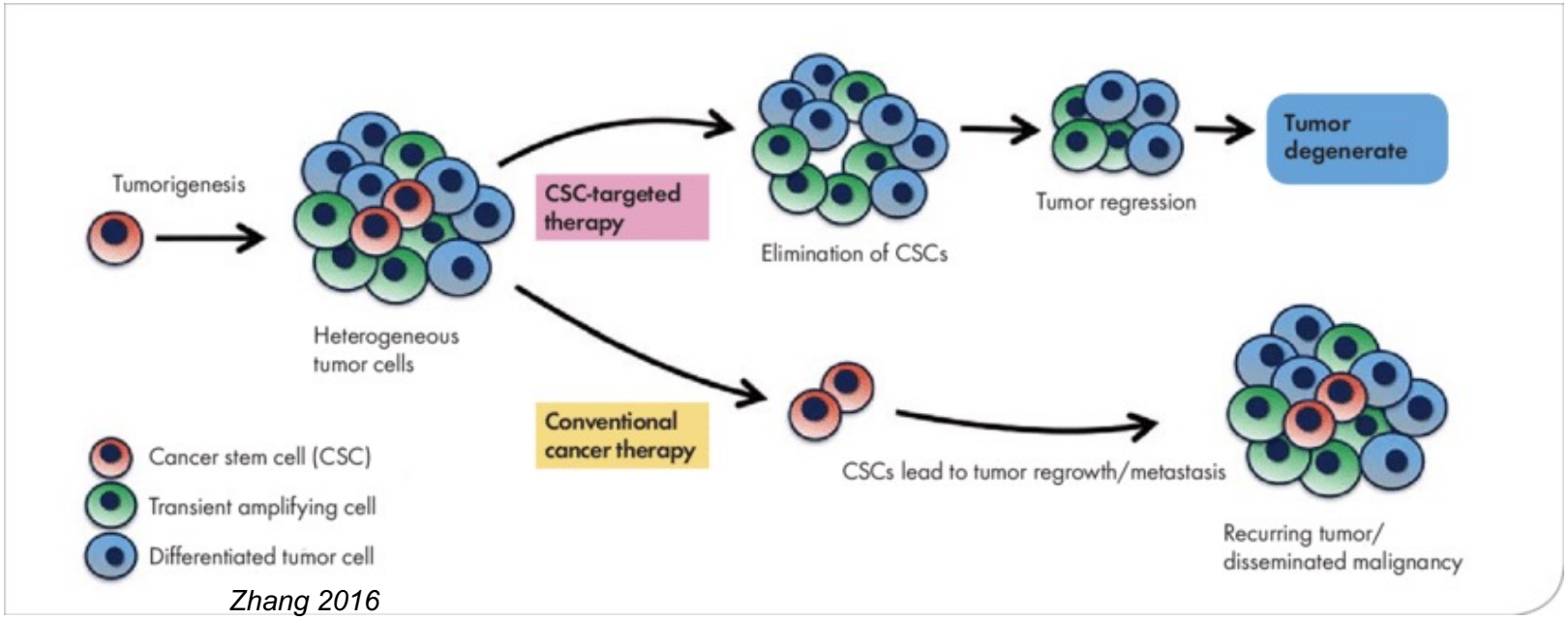


Zhang 2016

- The cancer stem cell model postulates that cancer cells form a hierarchy similar to the one of tissue stem cells, progenitors, and specialized cells

Cancer stem cell model - implications for therapy

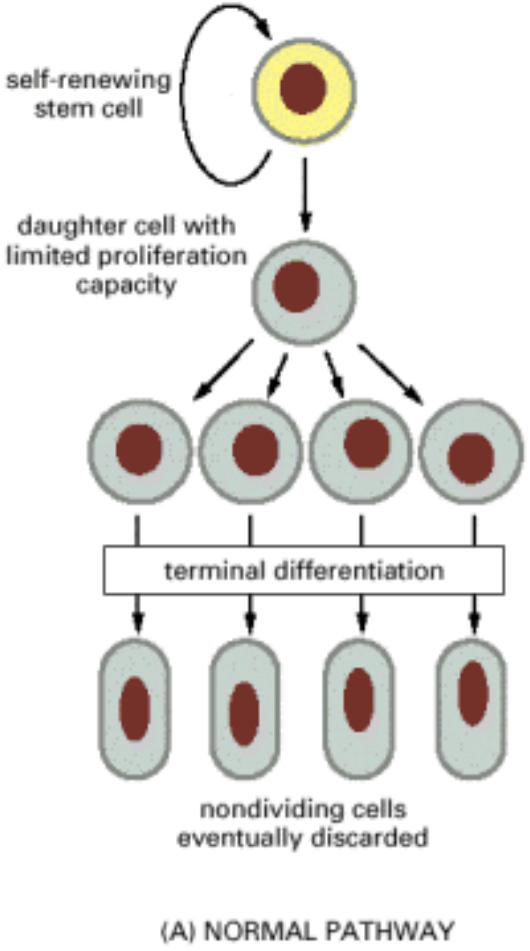
- See lecture “Cancer stem cells” by Prof. Radtke



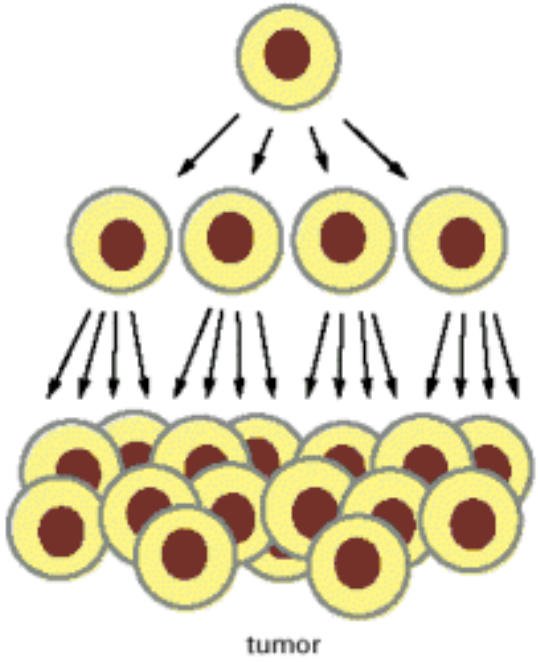
Zhang 2016

- CSCs are prognostic markers
- Often resistant to classical therapies and can cause relapses
- New therapies aim at specifically targeting them

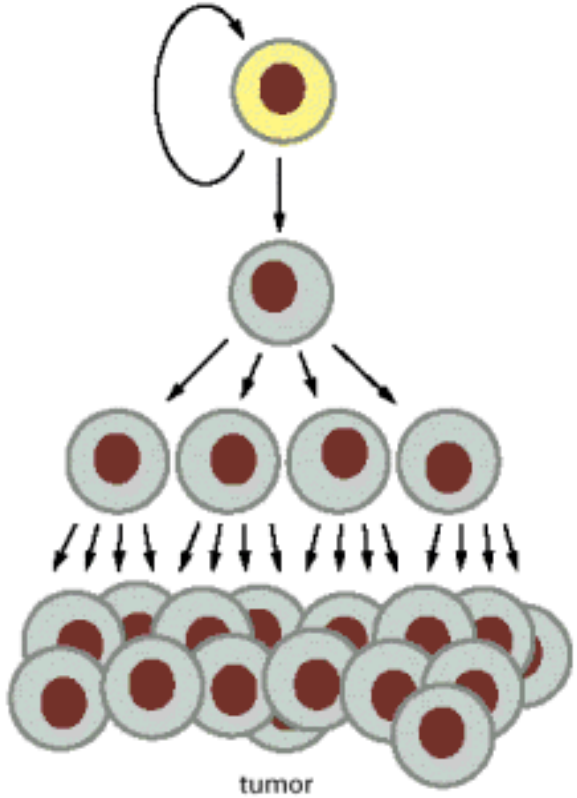
Balancing self-renewal and differentiation is crucial



(A) NORMAL PATHWAY



(B) STEM CELL FAILS TO PRODUCE ONE NON-STEM-CELL DAUGHTER IN EACH DIVISION AND THEREBY PROLIFERATES TO FORM A TUMOR

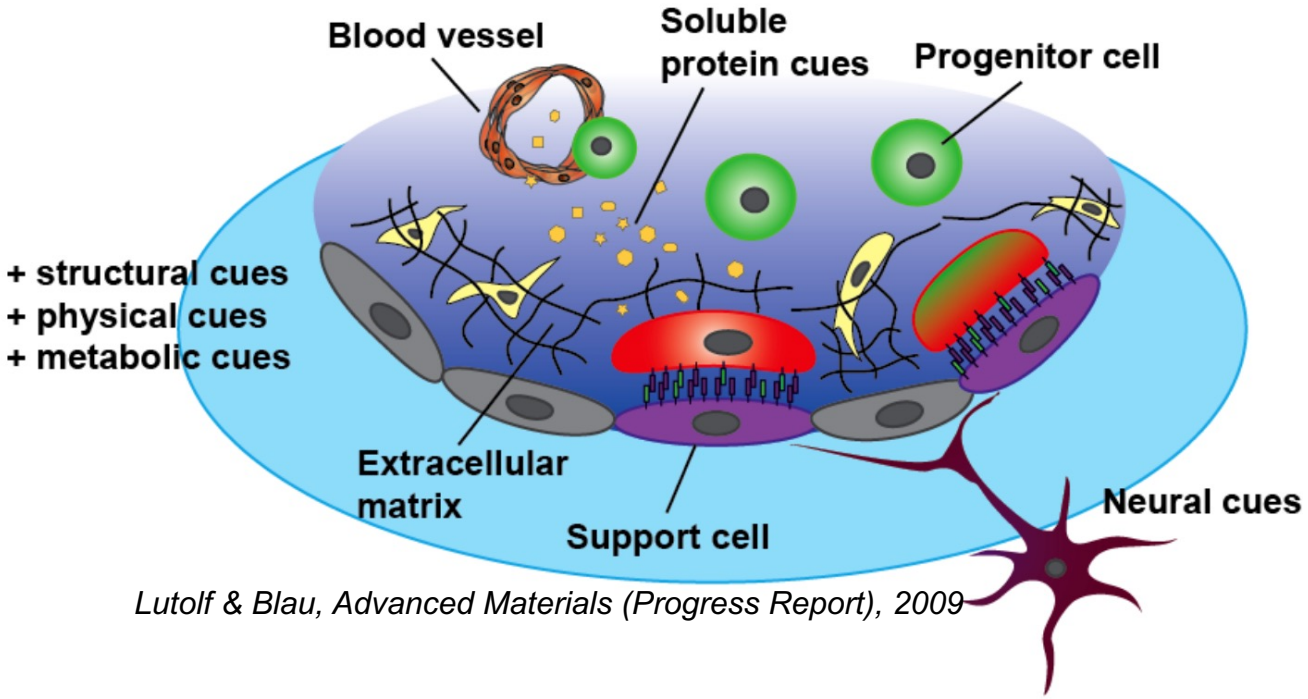


(C) DAUGHTER CELLS FAIL TO DIFFERENTIATE NORMALLY AND INSTEAD PROLIFERATE TO FORM A TUMOR

How is this balance regulated ?

The stem cell niche

- See lecture “Stem cell niche” by Prof. Karthaus

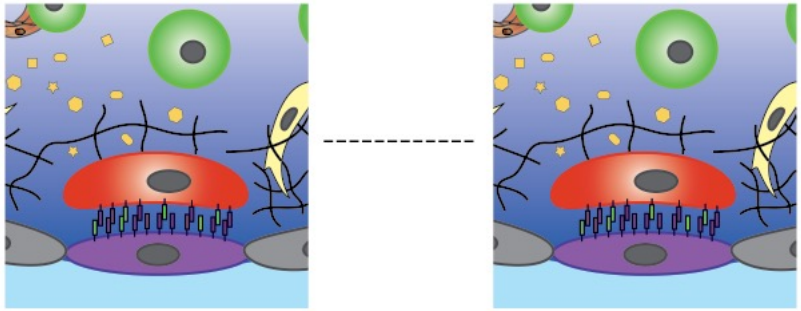


Lutolf & Blau, Advanced Materials (Progress Report), 2009

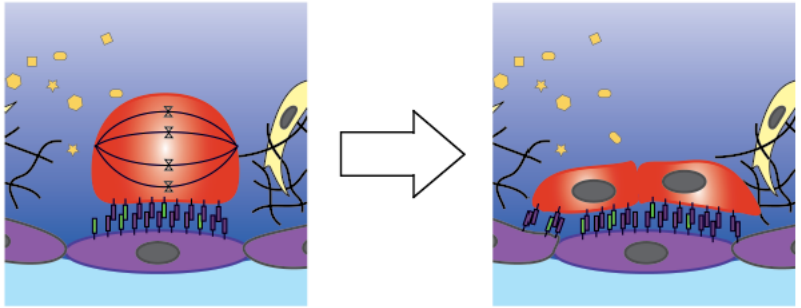
- Defined location and architecture
- Biochemical stem cell-niche crosstalk
- Physical interaction with support cells
- Protects stem cells from differentiation
- Regulates their renewal

Possible stem cell “fates” in the niche

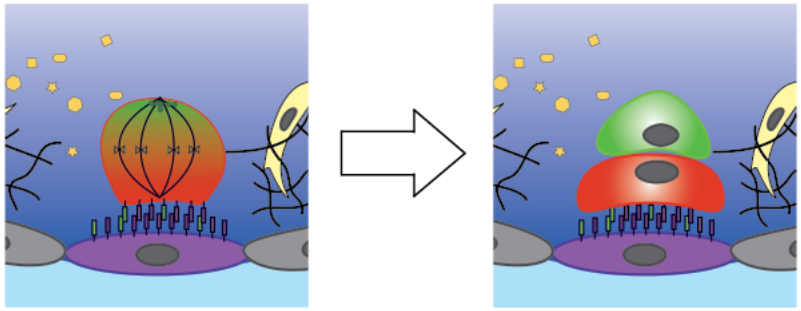
Quiescence



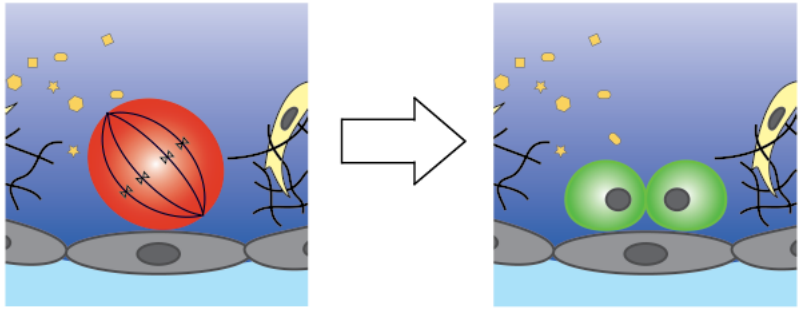
Symmetric self-renewal division



Asymmetric self-renewal division



Differentiation division



What makes stem cells so interesting?

Stem cells play crucial roles

Stem cells play crucial roles during:

- Embryogenesis
- Maintenance of regenerative tissues such as skin, intestine and the hematopoietic system
- Tissue repair after injury (skin, muscle, bone, gut, …)
- Cancer (Cancer stem cells)

→ Many potential clinical applications

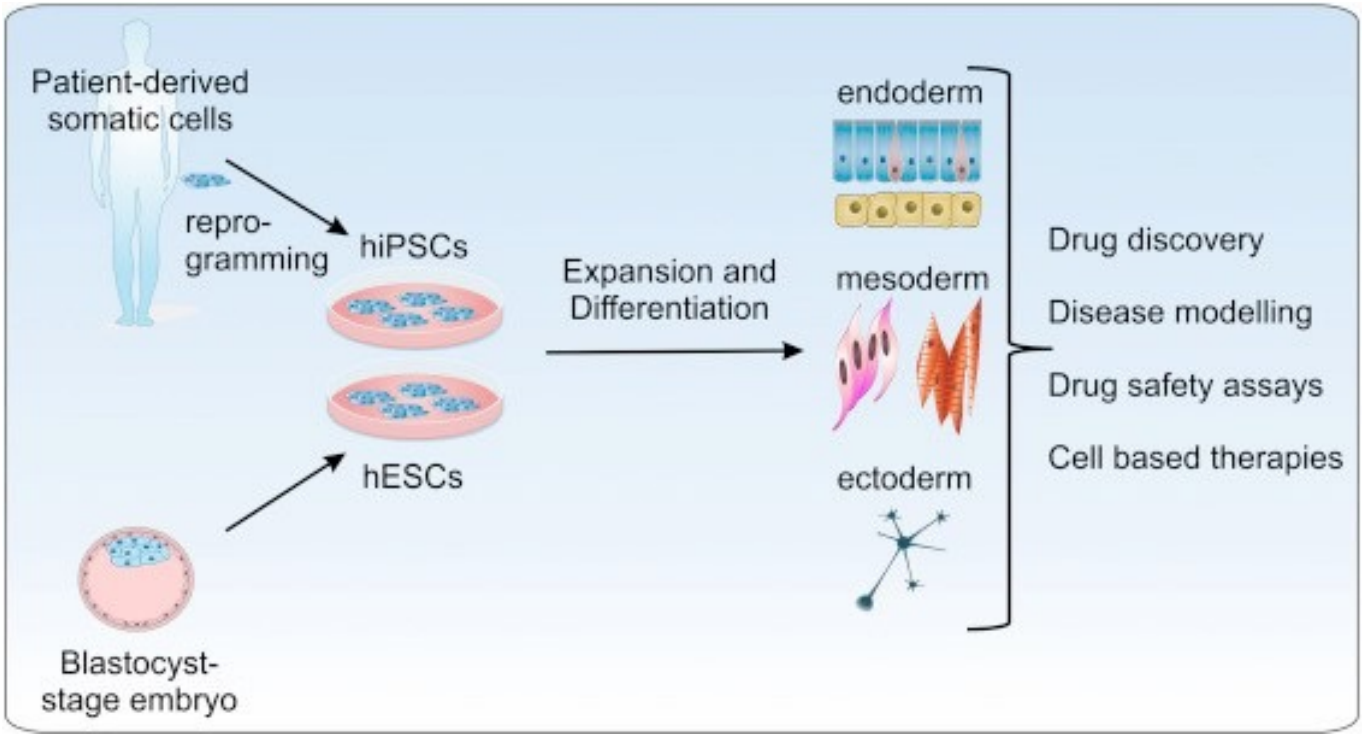
Stem cell - based therapy

- Differentiation of stem cells to produce tissues to replace
  - hESCs
  - iPSCs
  - Tissue stem cells in vitro
- Gene therapy on stem cells
- Disease modelling, drug screenings

Stem cell - based disease modelling

- In vitro disease model
- Drug discovery
- Drug safety assays

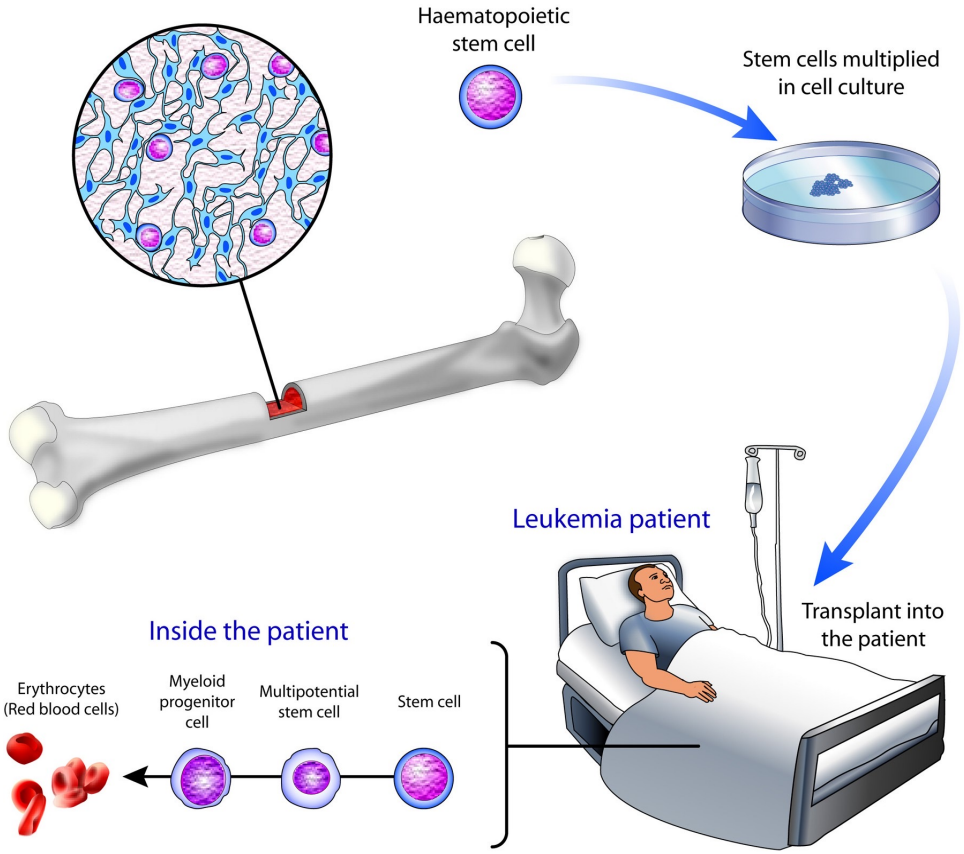
- See lectures “Clinical application” by Prof. Naveiras



Kropp et al., Process biochem. 2017

Stem cell based therapy – an example

- Bone marrow transplantation



EBMT Activity Survey in 2018: Patient and transplant numbers

Indication	Allogeneic HSCT	Autologous HSCT	Total
1st allo/1st auto HSCT	18,483	24,418	42,901
Additional HSCT	1,147	3,420	4,567
TOTAL	19,630	27,838	47,468

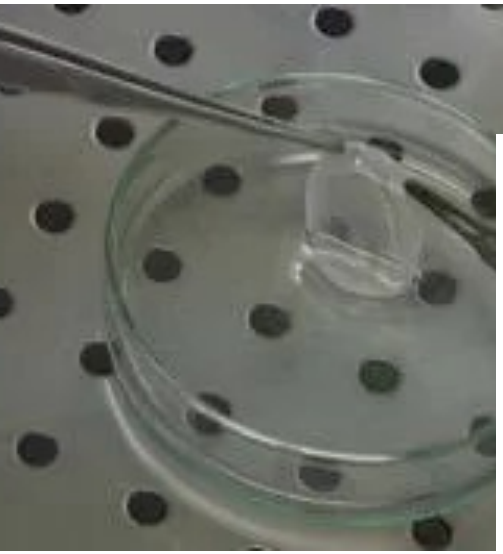
Teams: 701(of 720) Countries reporting: 50

EBMT Activity Survey 2018: Main indications

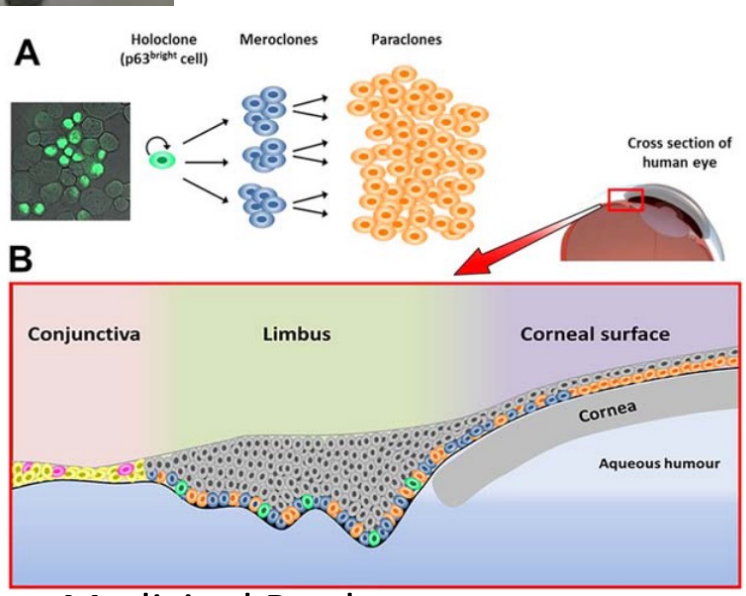
Indication	Allogeneic 1st HSCT	Autologous 1st HSCT	Total
Myeloid (AML, CML, MDS/MPN)	10,385	294	10,679
Lymphoid (ALL, CLL, HL, NHL)	4,953	8,803	13,756
Plasma cell disorder	416	13,146	13,562
Solid tumor	47	1,578	1,625
Non-malignant disorders	2,487	576	3,063
bone marrow failure	*954*	*5*	*959*
auto immune disease	*20*	*550*	*570*
Other	195	21	216
Total Patients	18,483	24,418	42,901

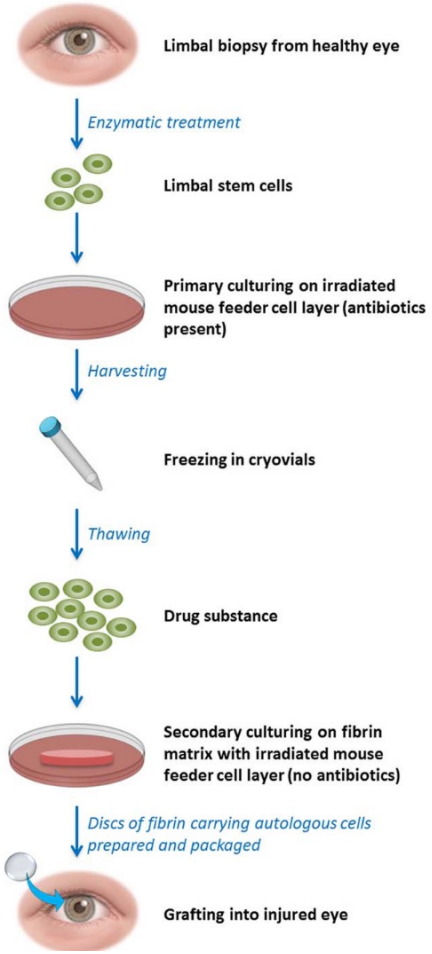
Stem cell-based therapy – first ATMP containing stem cells

- Holoclar is the first EMA-approval as first advanced therapy medicinal product containing stem cells in Europe
- To replace damaged cells on the surface of the cornea



https://www.eurostemcell.org/de





ATMP: Advanced Therapy Medicinal Product
EMA: European Medicines Agency

Pellegrini et al. Stem cells Transl. medicine 2018

Stem cells open questions/hurdles

- Very rare (dispersed and virtually invisible)
- Markers?? (with exception of the HSCs) → purity??
- *In vivo* and *in vitro* systems to assay SC activity
- Difficulty to culture stem cells in vitro (ESCs and iPS: differentiation, TSCs: self-renewal)
- In vivo use: survival?, rejection?, tumor formation?

A Stem Cell Story

https://www.youtube.com/watch?v=2-3J6JGN-_Y

Next lecture 20.09
(Stem) Cell Plasticity