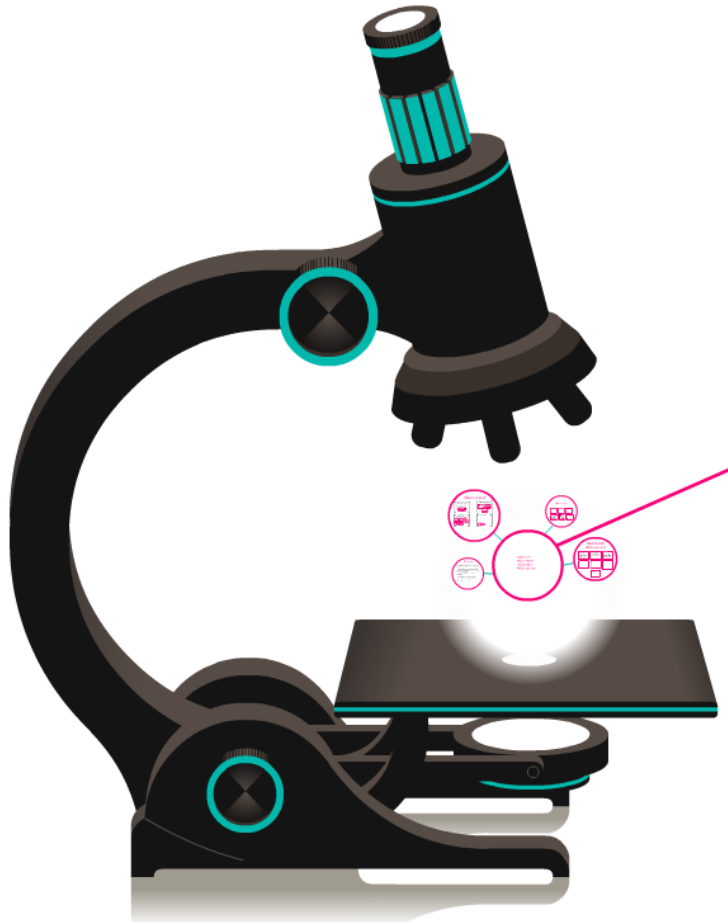


Stem Cells

Definition?
Where from?
What's for?
Where are we?

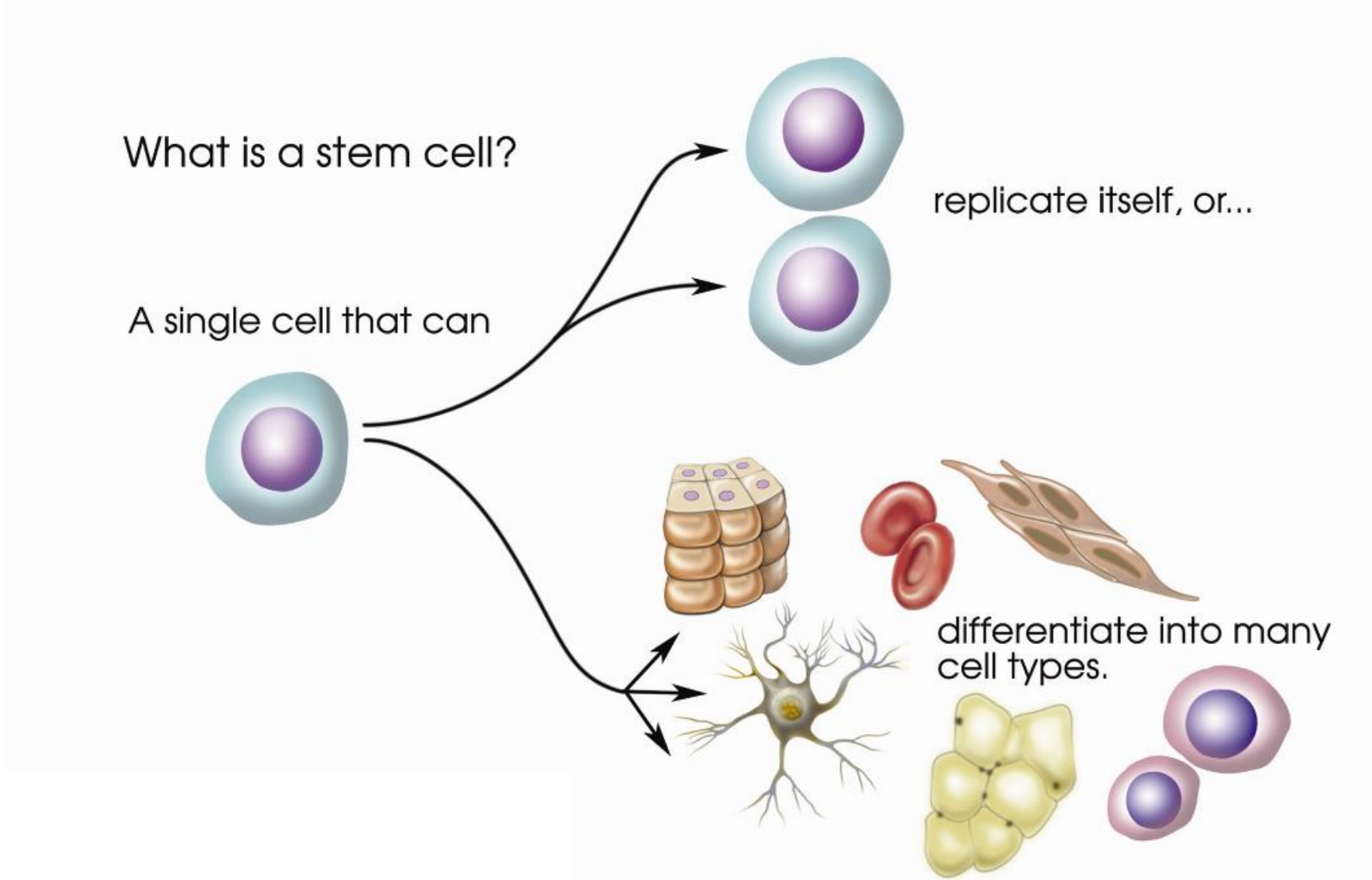
Definition

It is now well accepted that a stem cell must fulfill three criteria:

1- it must be capable of self-renewal, i.e., undergoing symmetric or asymmetric divisions through which the stem cell population is maintained.

2- A single cell must be capable of multilineage differentiation.

3- In vivo functional reconstitution of a given tissue.


What is a stem cell?
A single cell that can
replicate itself, or...
differentiate into many cell types.

Stem cells (SCs) are very small cells that do not have the phenotypic characteristics of cells from any known adult tissue (epithelial, connective, muscle, neural, and immune) but are able to generate ‘de novo’ differentiated cells of the types found in any of these tissues.

The definition of ‘stem cell’ is essentially functional: “rather than referring to a discrete cellular entity, a stem cell most accurately refers to a biological function that can be induced in many distinct types of cells, even differentiated cells”

STEM CELLS

- Embryonic stem cells (ESC)
- Adult stem cells (ASC)

(**physiological** stem cells that are present at different stages of the life)

- Nuclear transfer stem cells (NTSC)
- Induced pluripotent stem cells (iPS)

(**artificially reprogrammed** stem cells, by nuclear transfer or "induction")

- Cancer stem cells (CSC)

(**pathological** stem cells that are present in cancer)

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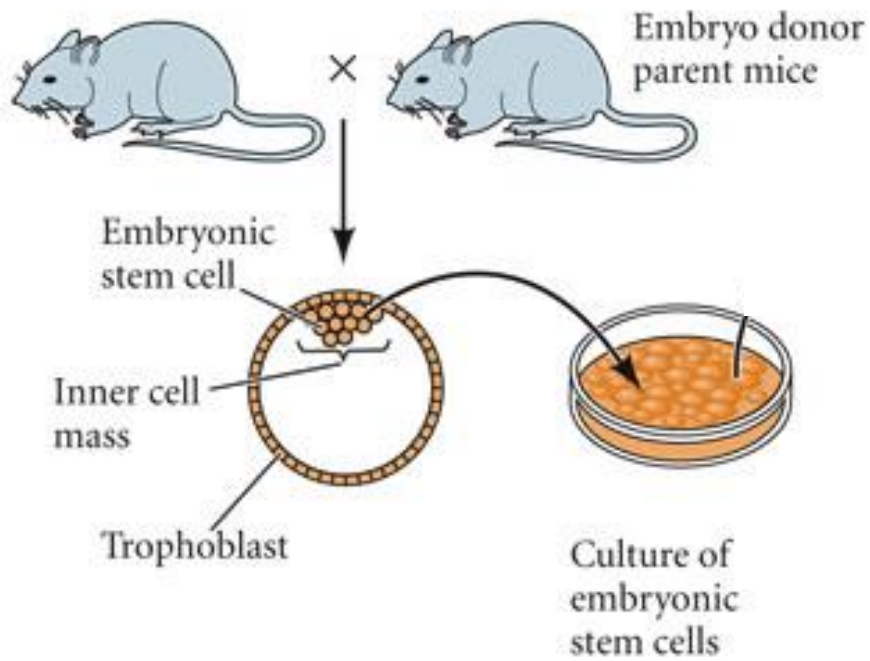
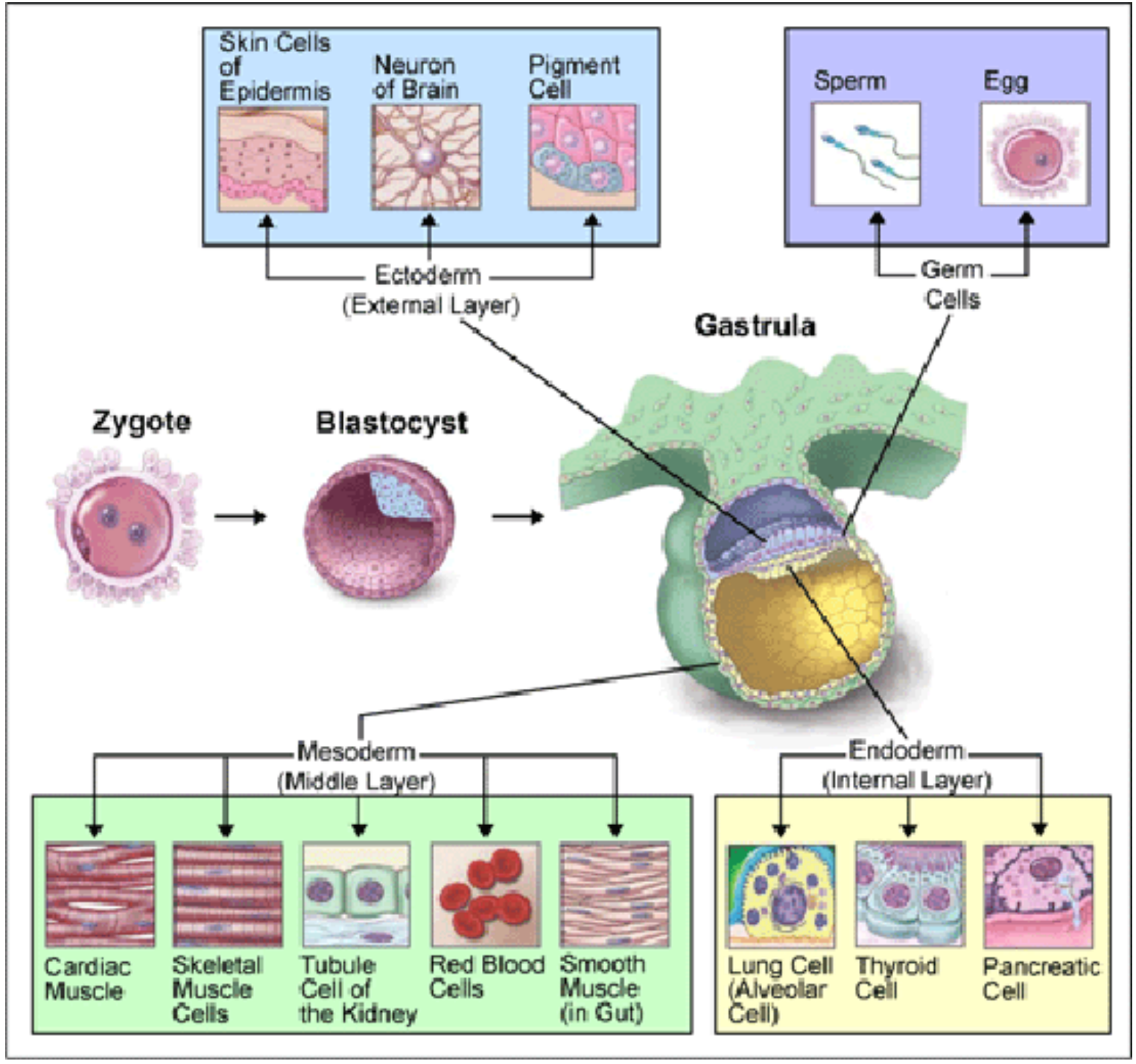
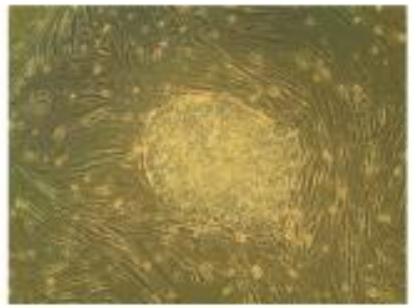
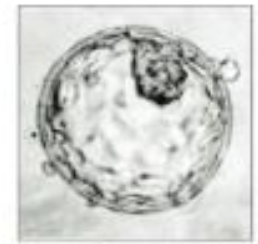
Embryonic stem cells (ESCs)

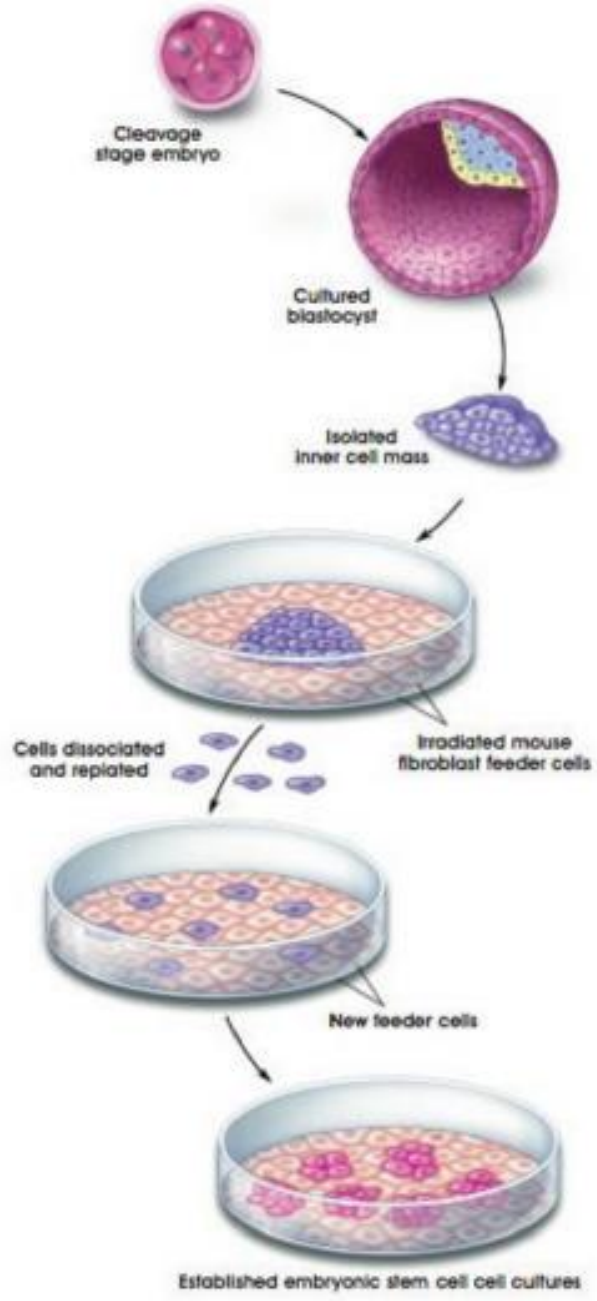
ESCs make up the inner cell mass (ICM) of the blastocyst before implantation or before any commitment to embryonic cell fates is detectable at the molecular level. They are the most studied SCs and knowledge obtained from ESCs has guided the investigations of other types of SC.




Skin Cells of Epidermis
Neuron of Brain
Pigment Cell
Sperm
Egg
Ectoderm (External Layer)
Germ Cells
Gastrula
Zygote
Blastocyst
Mesoderm (Middle Layer)
Endoderm (Internal Layer)
Cardiac Muscle
Skeletal Muscle Cells
Tubule Cell of the Kidney
Red Blood Cells
Smooth Muscle (in Gut)
Lung Cell (Alveolar Cell)
Thyroid Cell
Pancreatic Cell

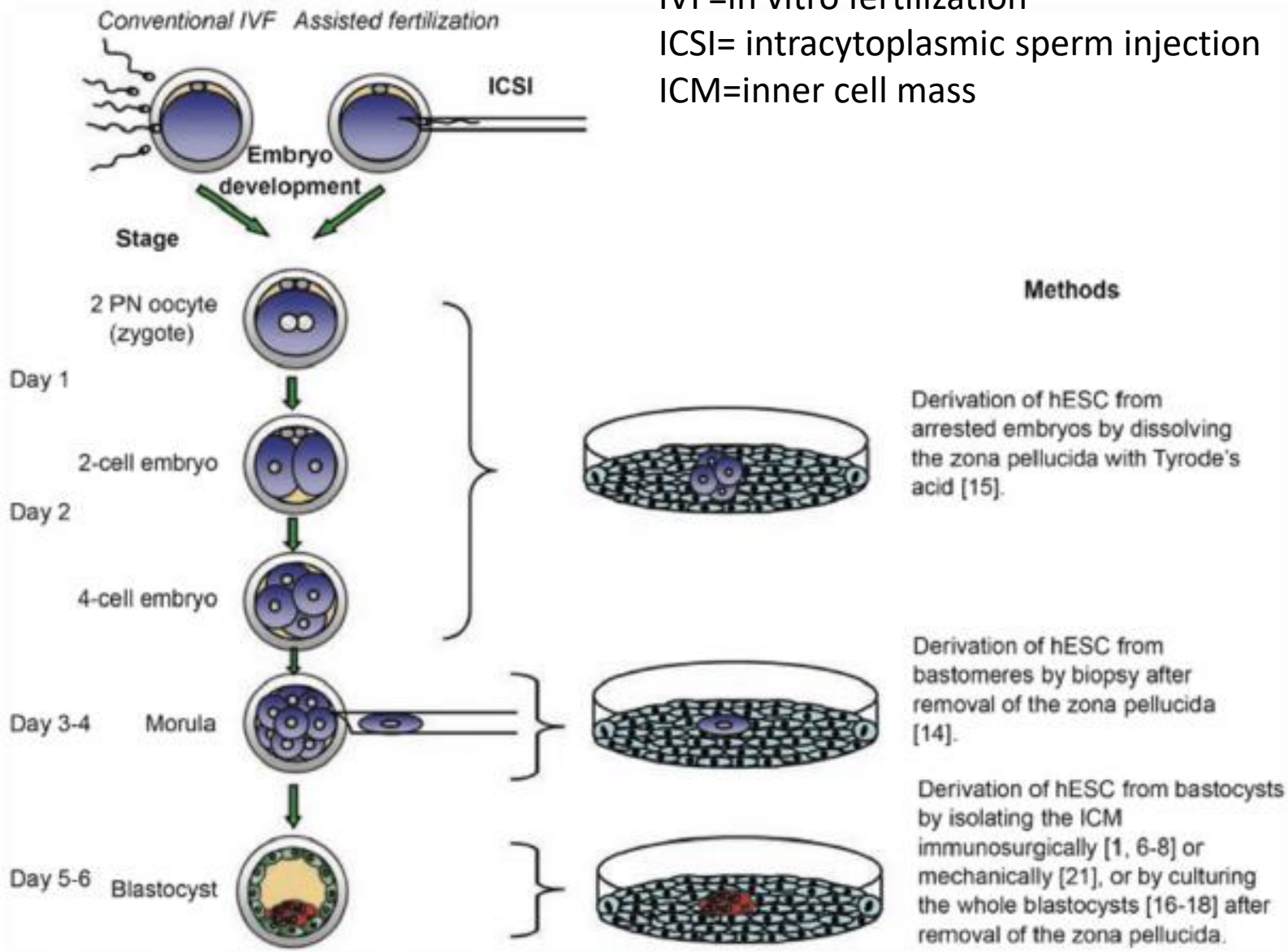


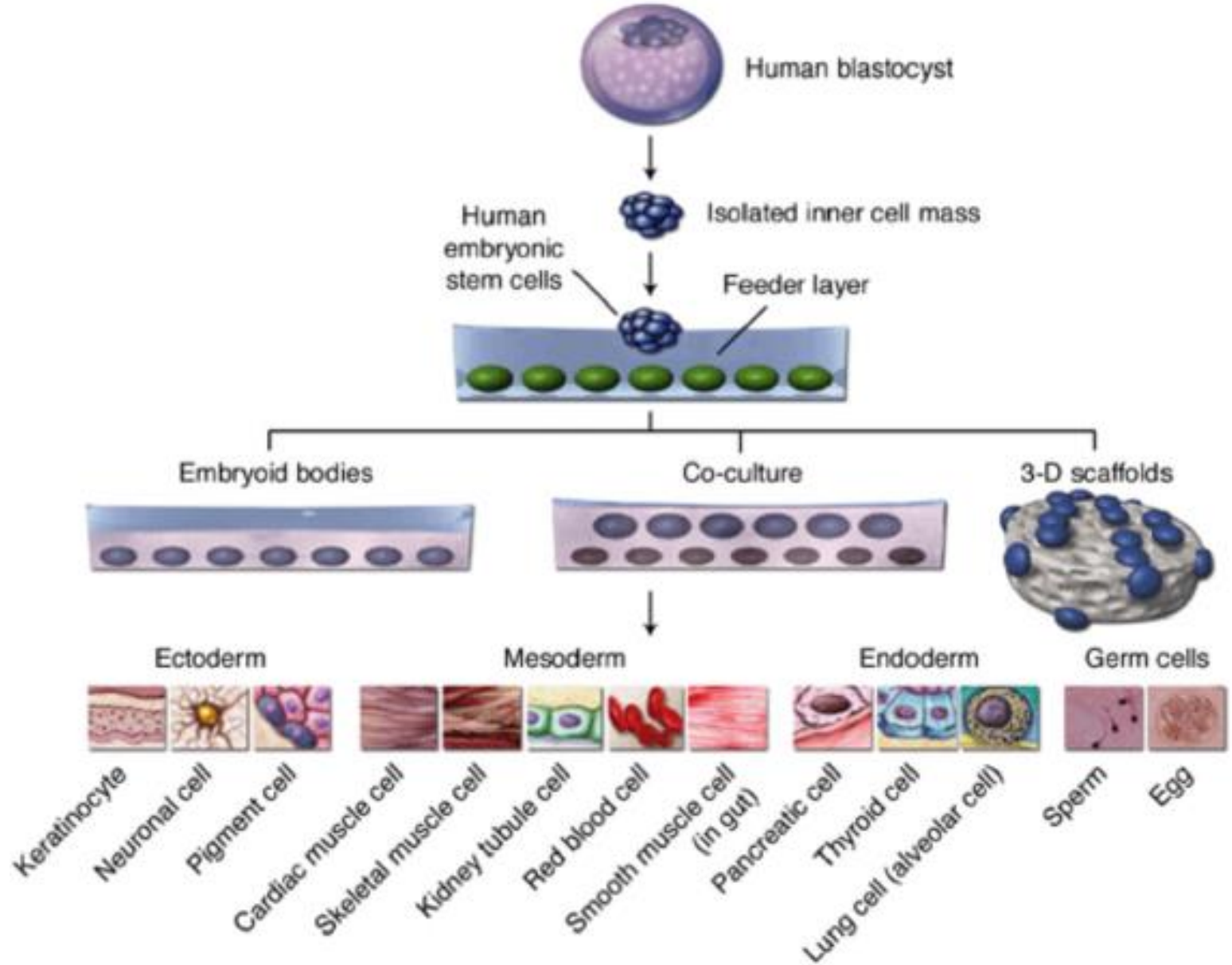



Cleavage stage embryo
Cultured blastocyst
Isolated inner cell mass
Irradiated mouse fibroblast feeder cells
Cells dissociated and replated
New feeder cells
Established embryonic stem cell cell cultures

In human

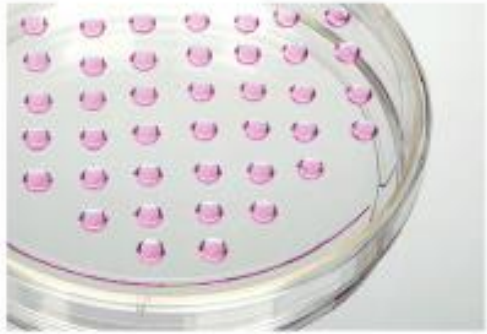
IVF=in vitro fertilization
ICSI= intracytoplasmic sperm injection
ICM=inner cell mass



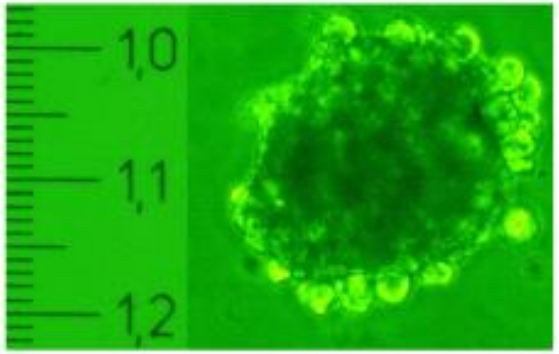


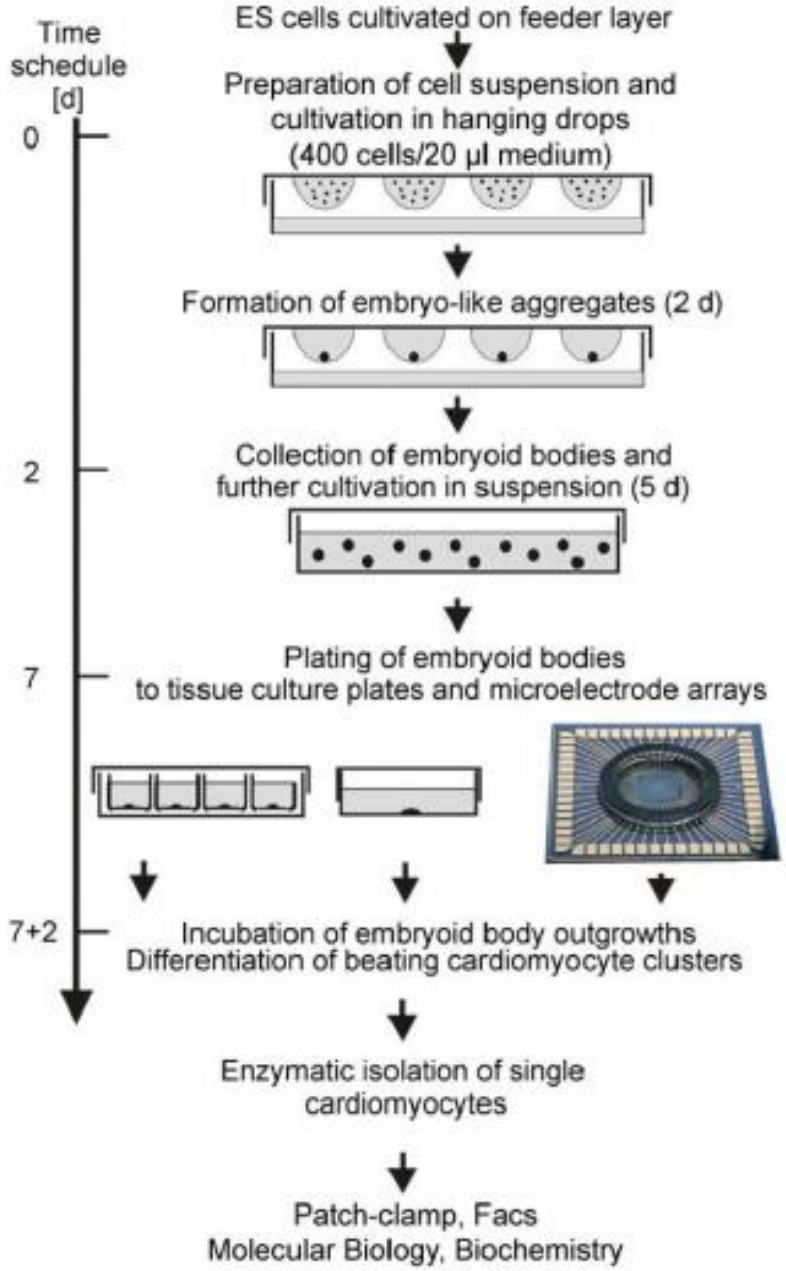
Derivation of a human embryonic stem cell line, and differentiation strategies

2nd step:
Embryoid bodies

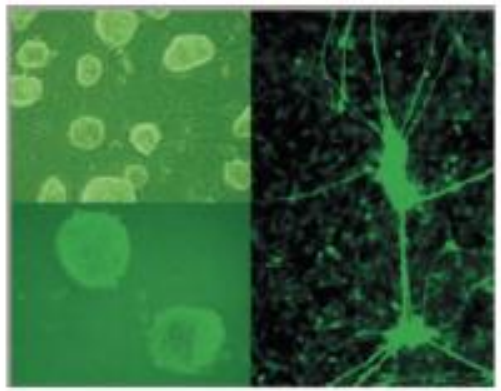


Hanging drop culture. Embryoid bodies for the differentiation of embryonic stem cells in the embryonic stem cell test are generated by pipetting a single-cell suspension onto the lid of a cell culture dish. The cells aggregate at the bottom of the drop by gravitational force, thereby forming the embryoid body.

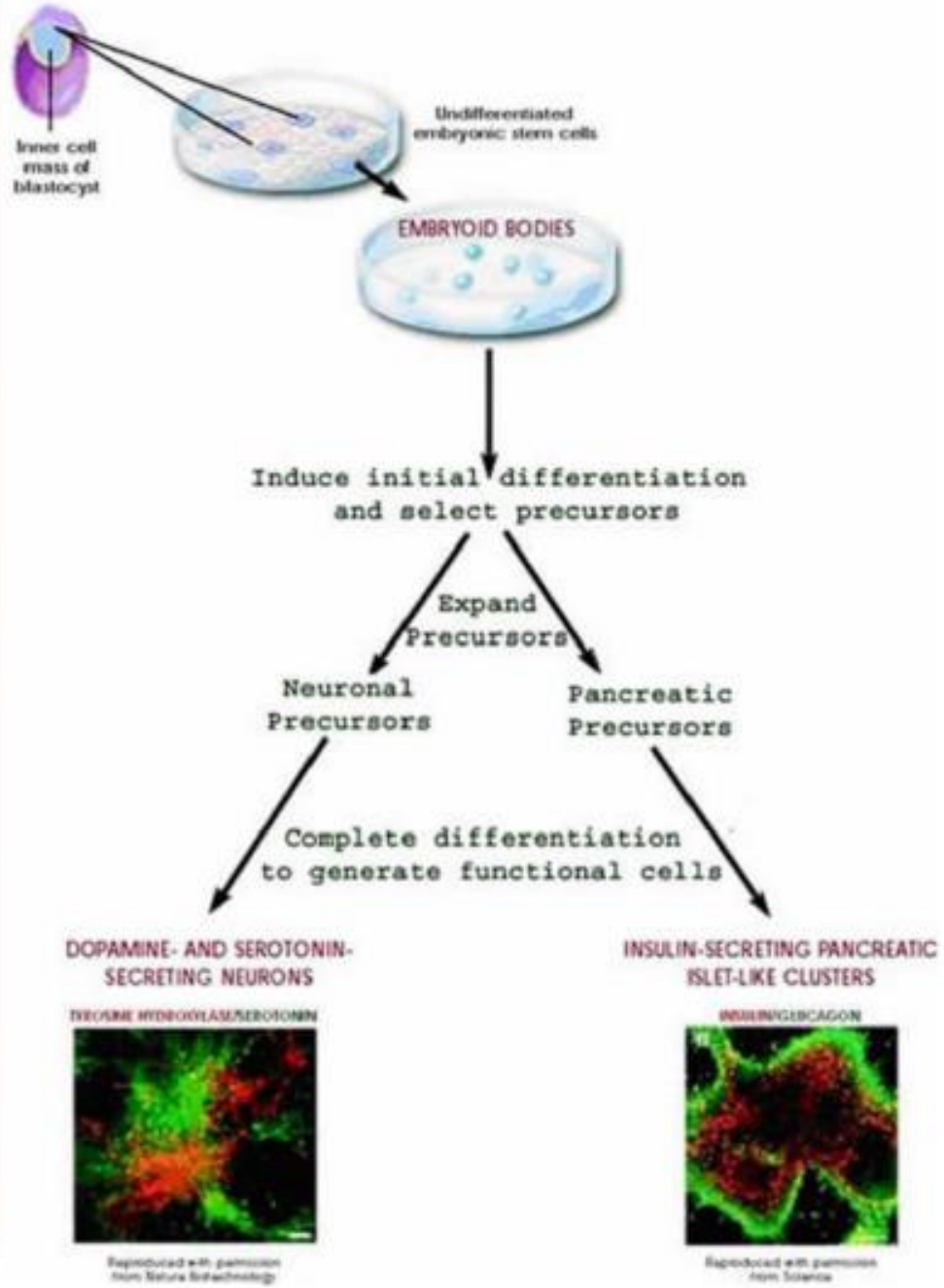




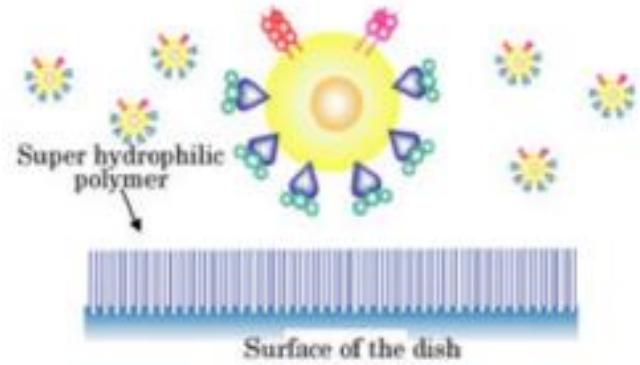
3rd step: terminal differentiation



A nerve cell (the right) derived from mouse undifferentiated cells (the top left) via an embryoid body (an assembly of the undifferentiated cells, the bottom left) in vitro.

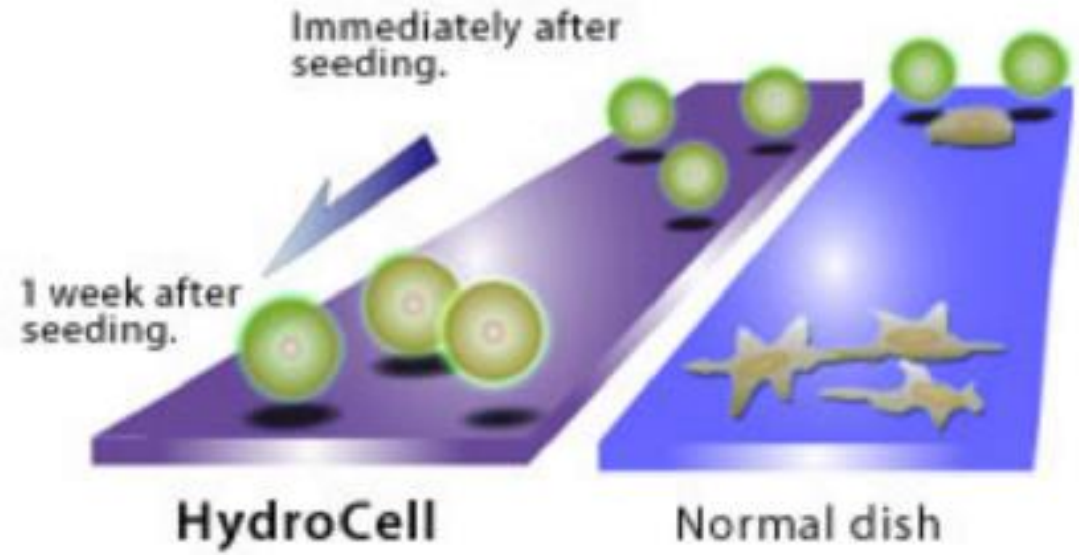


Alternative to hanging drop:
Low cell binding plates/dishes HydroCell



Key features of HydroCell®

♦Super hydrophilic polymer is fixed to the surface of the dish/plate at nano-thickness level.
♦The surface is highly cells/protein resistent
♦Applications
Improved macrophage culture "in vitro".
Embryoid body formation from ES cells.
Spheroid formation in culture.



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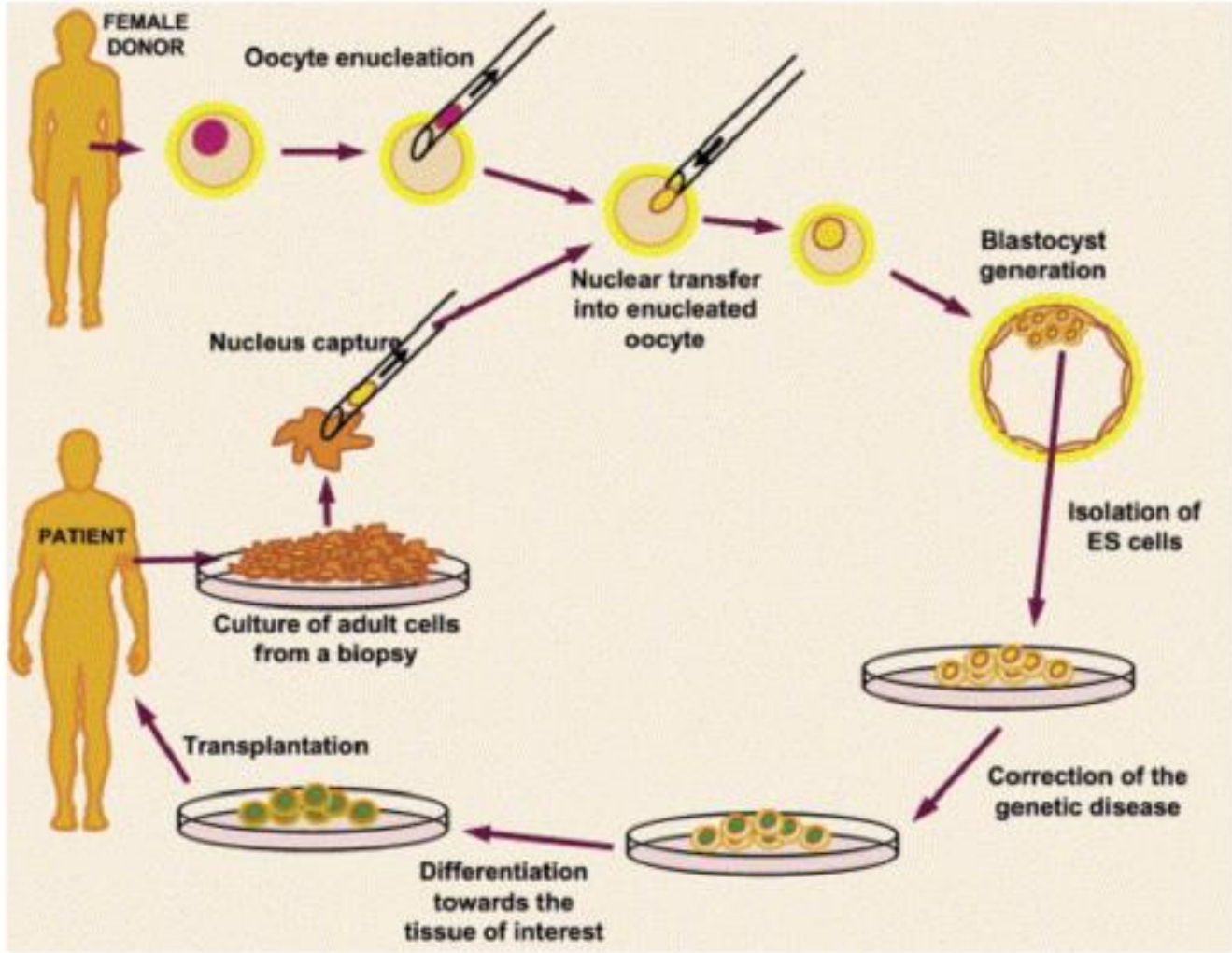
Embryonic stem cells obtained by nuclear transfer

Embryonic stem cells obtained by nuclear transfer are genetically identical to the patient ---> no immune



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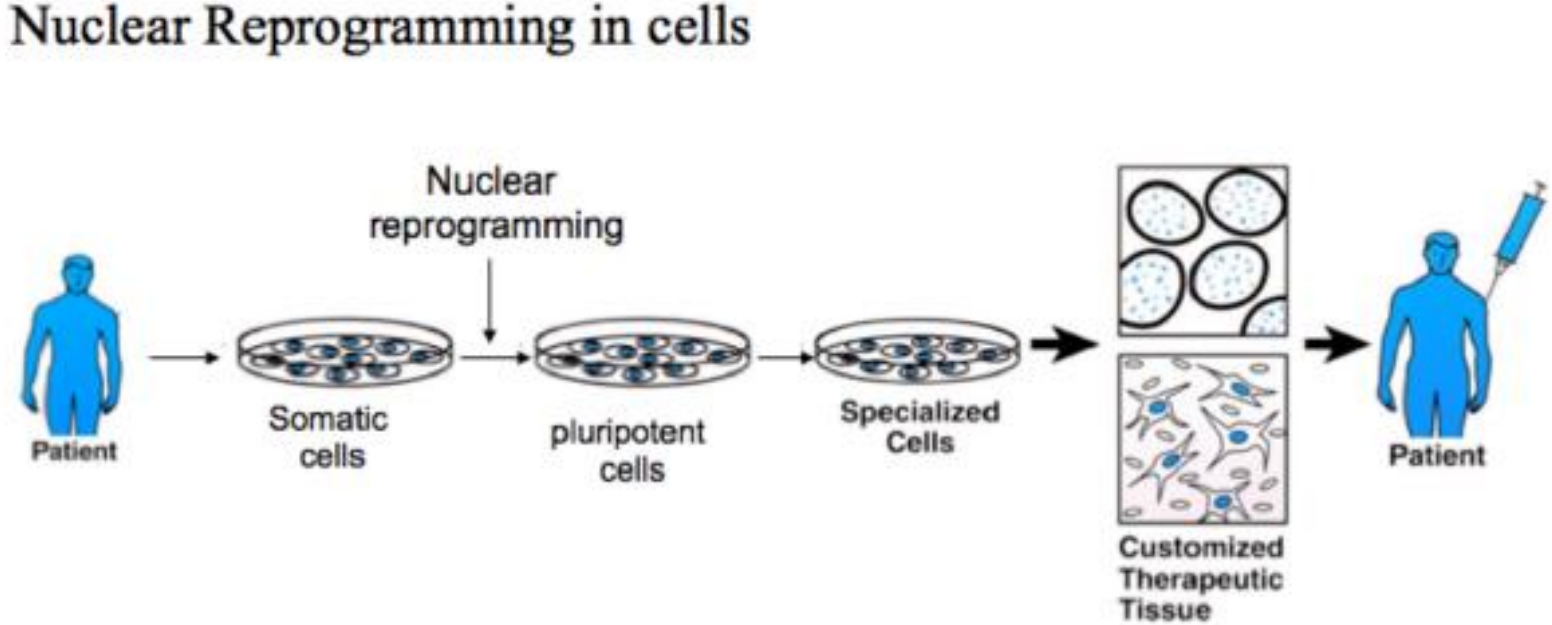
Induced pluripotent stem cell from adult cell o transdifferentiation



Cell *126*, 663–676, August 25, 2006

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

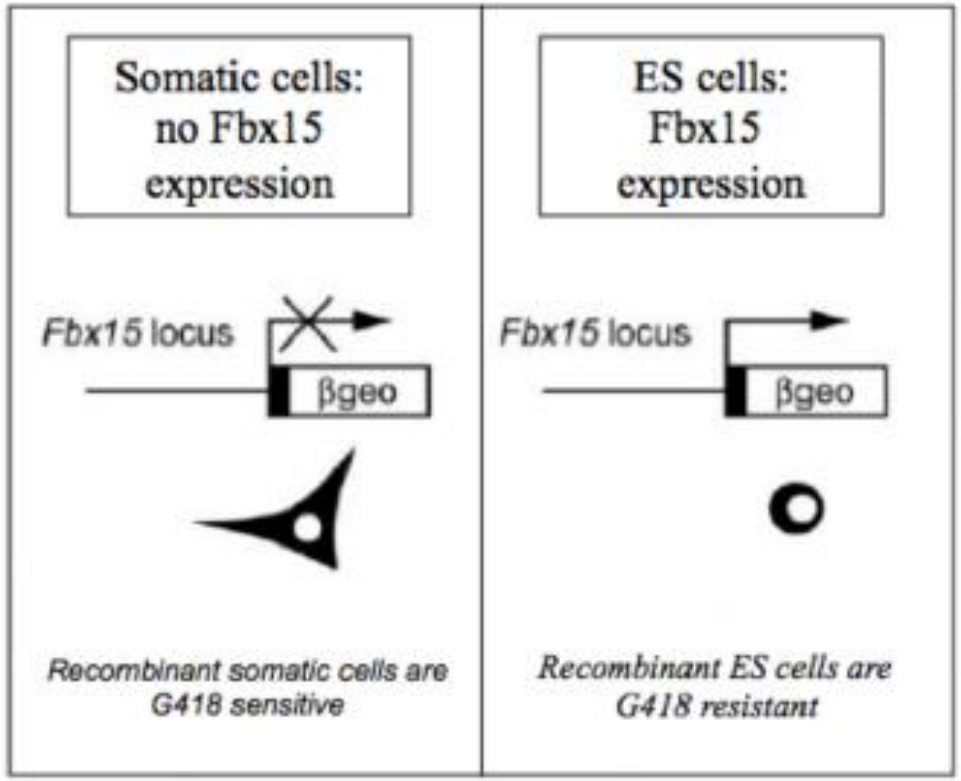
"We hypothesized that the factors that play important roles in the maintenance of ES cell identity also play pivotal roles in the induction of pluripotency in somatic cells".

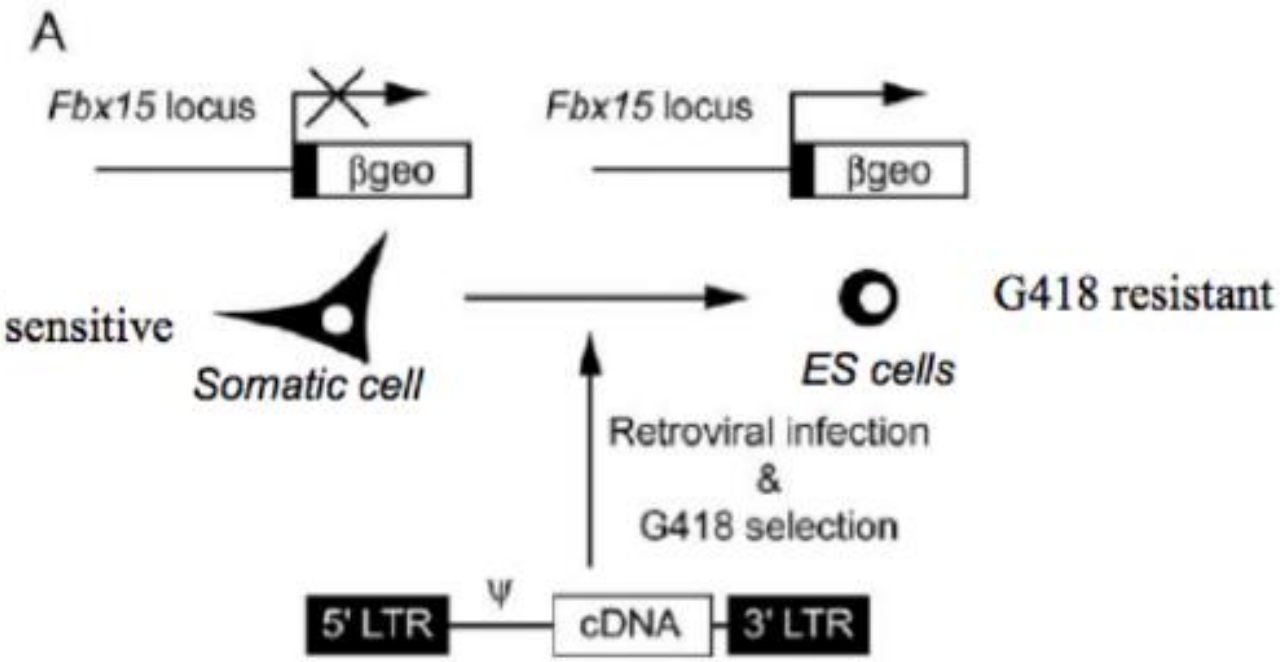
We selected 24 genes as candidates for factors that induce pluripotency in somatic cells, based on our hypothesis that such factors also play pivotal roles in the maintenance of ES cell identity. For b-catenin, c-Myc, and Stat3, we used active forms, S33Y-b-catenin (Sadot et al., 2002), T58A-c-Myc (Chang et al., 2000), and Stat3-C (Bromberg et al., 1999), respectively.
Because of the reported negative effect of Grb2 on pluripotency (Burdon et al., 1999; Cheng et al., 1998), we included its dominant-negative mutant Grb2DSH2 (Miyamoto et al., 2004) as 1 of the 24 candidates.

Assay system in which the induction of the pluripotent state could be detected as resistance to G418.

We inserted a βgeo cassette (a fusion of the β -galactosidase and neomycin resistance genes) into the mouse Fbx15 gene by homologous recombination (Tokuzawa et al., 2003). Although specifically expressed in mouse ES cells and early embryos, Fbx15 is dispensable for the maintenance of pluripotency and mouse development.

ES cells homozygous for the β geo knockin construct (Fbx15 β geo/ β geo) were resistant to extremely high concentrations of G418 (up to 12 mg/ml), whereas **somatic cells** derived from Fbx15bgeo/bgeo mice were sensitive to a normal concentration of G418 (0.3 mg/ ml). We expected that even partial activation of the Fbx15 locus would result in resistance to normal concentrations of G418.

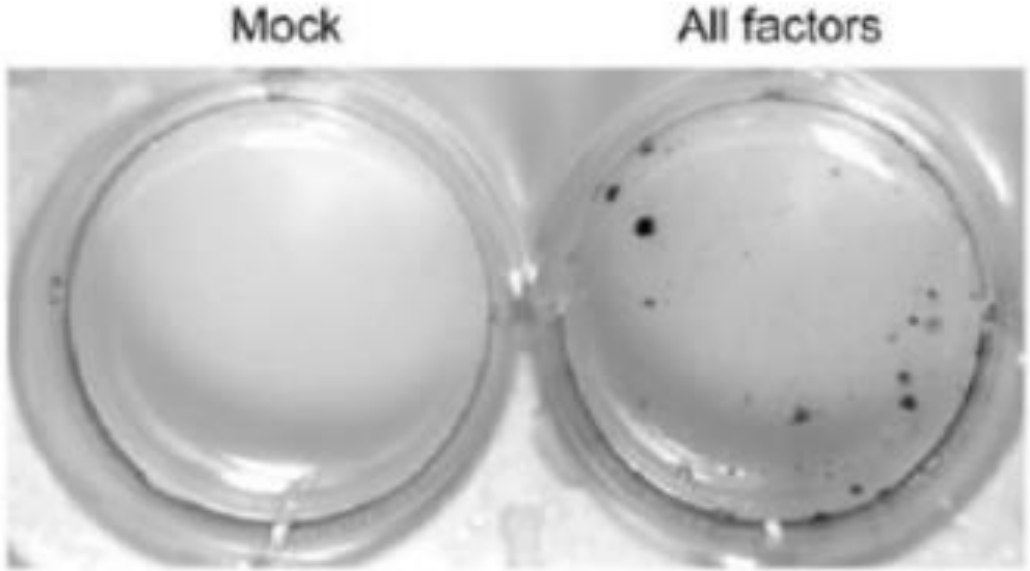




Retroviral infection with constructs for each of those factors known to be important for plutipotency

Ecat1	Klf4
Dppa5	β-catenin
Fbxo15	c-Myc
Nanog	Stat3
ERas	Grb2
Dnmt3l	Ecat8
Oct3/4	Gdf3
Sox2	Sox15
Rex1	Dppa4
Utf1	Dppa2
Tcl1	Fthl17
Dppa3	Sall4

We introduced each of the 24 candidate genes into mouse embryonic fibroblasts (MEFs) from Fbx15 β geo/ β geo embryos by retroviral transduction. Transduced cells were then cultured on STO feeder cells in ES cell medium containing G418 (0.3 mg/ml).

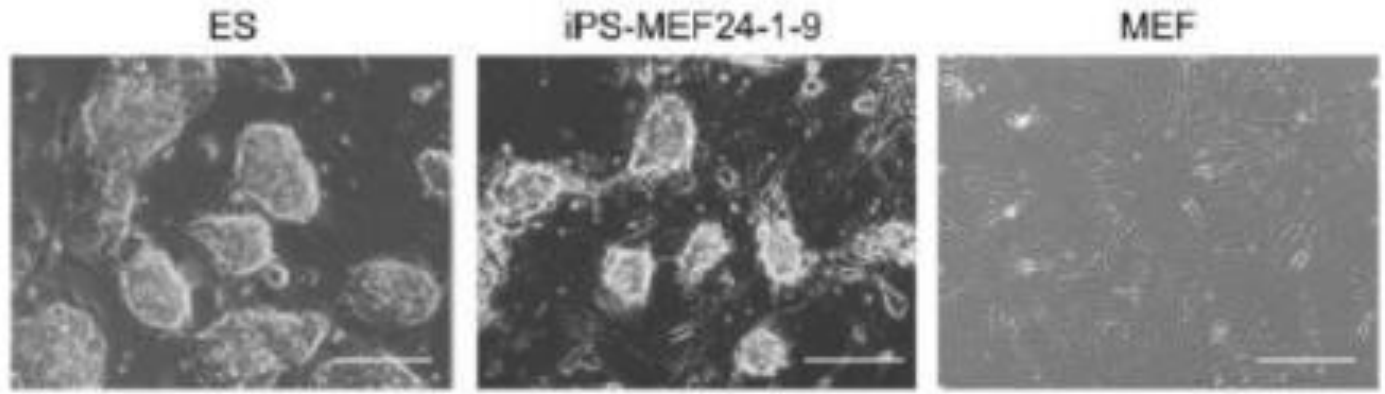


G418-resistant colonies were observed 16 days after transduction with a **combination of 24 factors**. Cells were stained with crystal violet.

no drug-resistant colonies with any single factor, indicating that no single candidate gene was sufficient to activate the Fbx15 locus

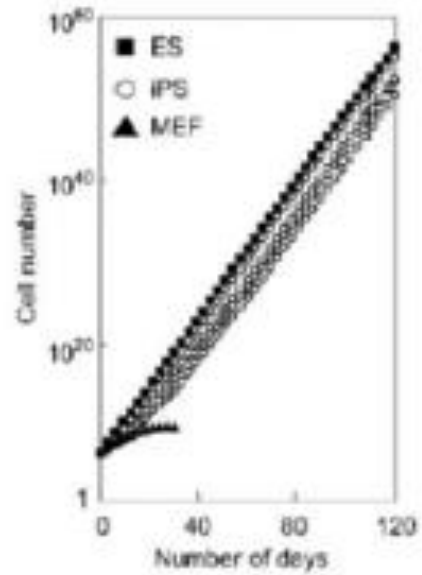
Of the 12 clones for which we continued cultivating under selection, 5 clones exhibited morphology similar to ES cells, including a round shape, large nucleoli, and scant cytoplasm

MES=mouse embryonic fibroblasts)



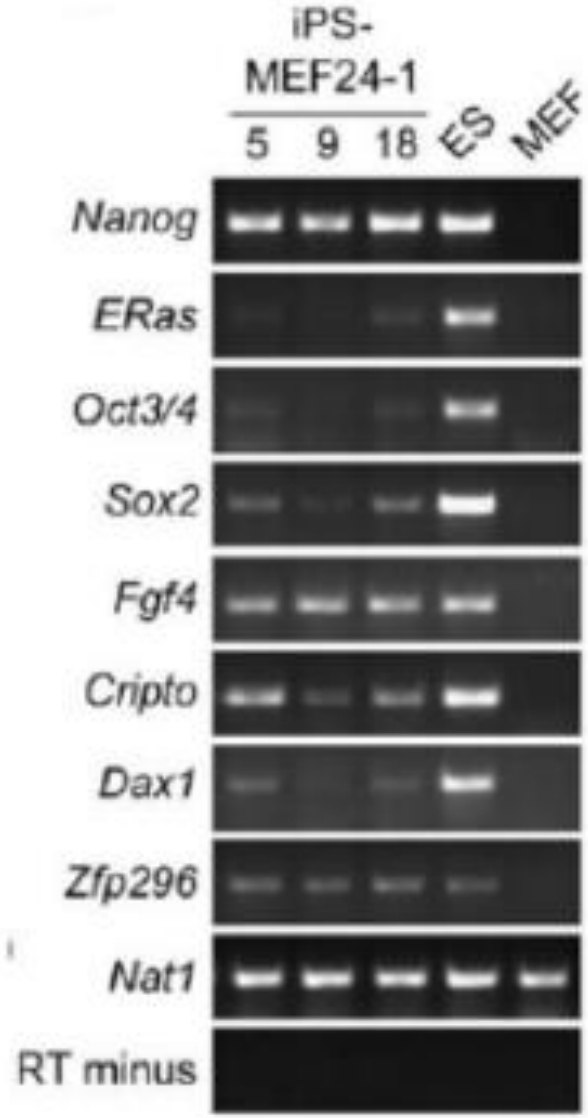
iPS-MEF24 = ''pluripotent stem cells induced from MEFs by 24 factors

4 of these clones possessed ES cell-like morphology and proliferation properties

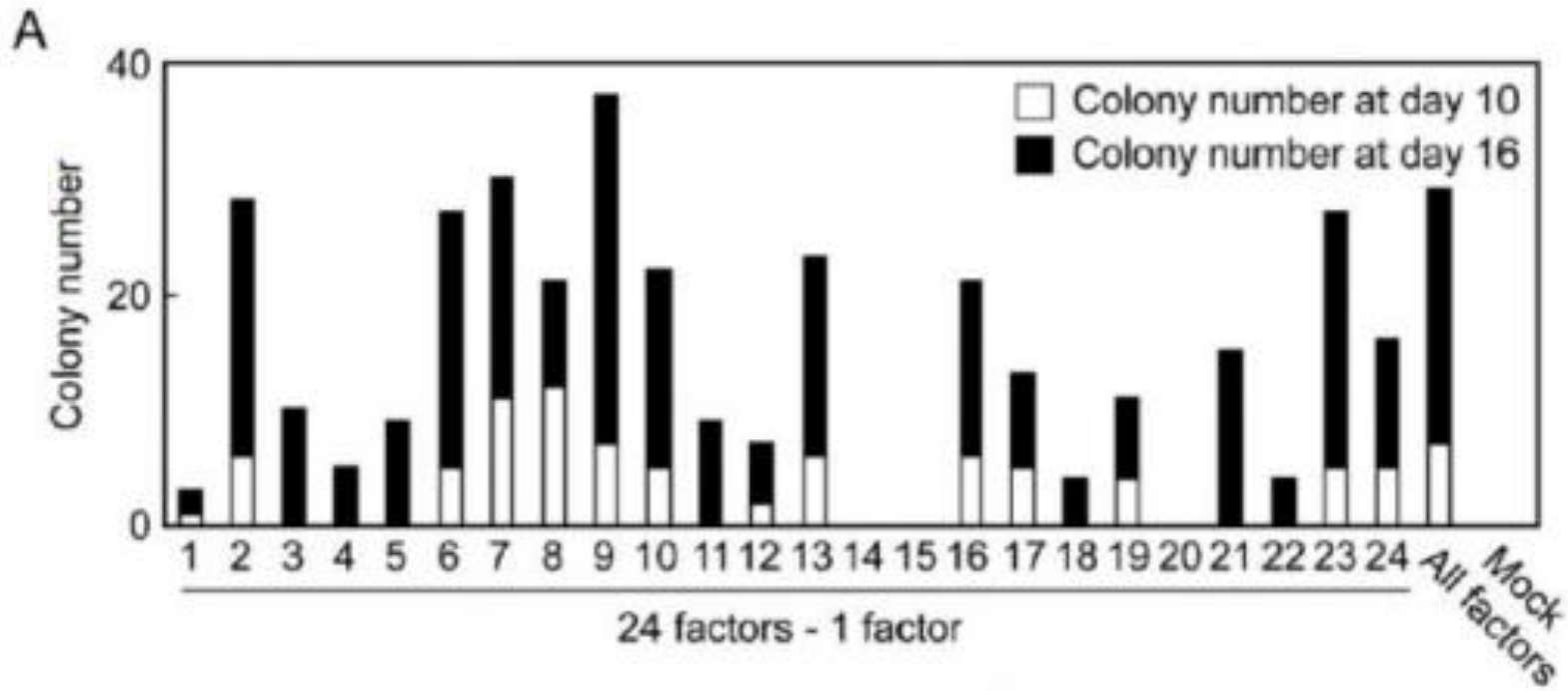


Reverse transcription PCR (RT-PCR) analysis revealed that the iPS-MEF24 clones expressed ES cell markers, including Oct3/4, Nanog, E-Ras, Cripto, Dax1, and Zfp296 and Fgf4

These data indicate that some combination of these 24 candidate factors induced the expression of ES cell marker genes in MEF culture.



we examined the effect of withdrawal of individual factors from the pool of transduced candidate genes on the formation of G418-resistant colonies



Effect of the removal of individual factors from the pool of 24 transduced factors on the formation of G418-resistant colonies. Fbx15bgeo/bgeo MEFs were transduced with the indicated factors and selected with G418 for 10 days (white columns) or 16 days (black columns).

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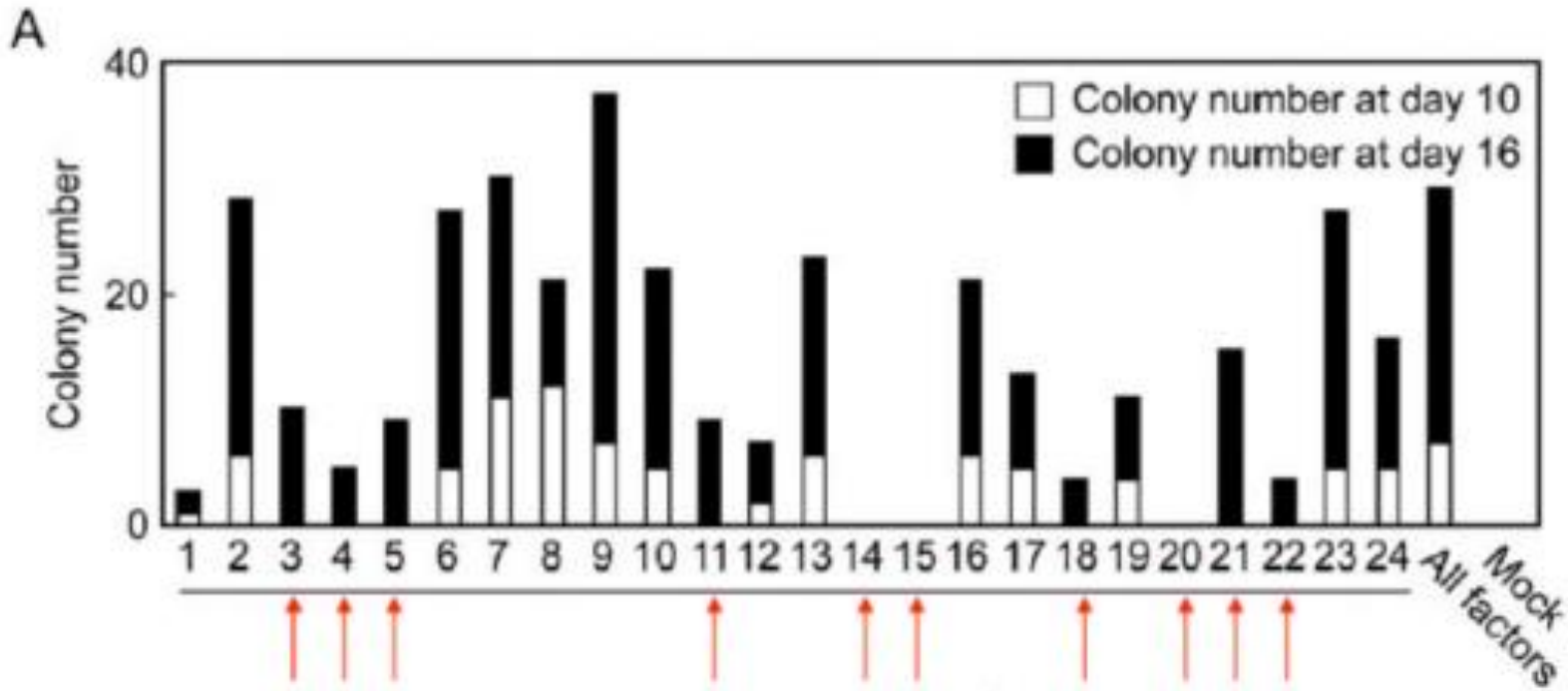
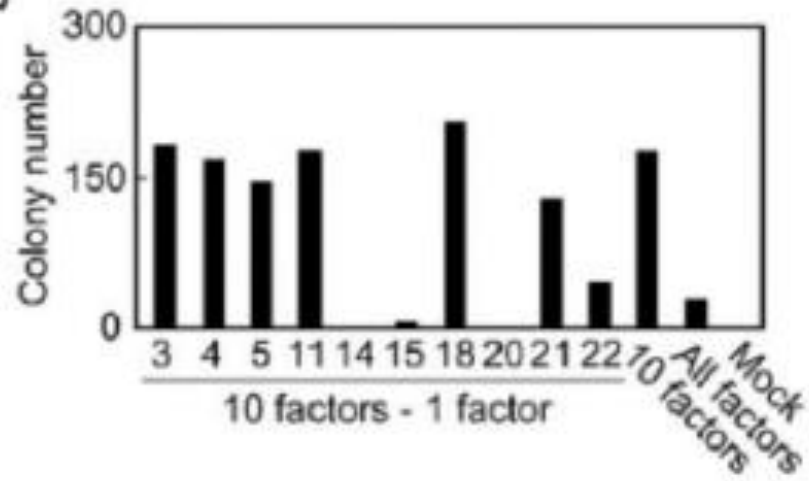


Effect of the removal of individual factors from the pool of 24 transduced factors on the formation of G418-resistant colonies. Fbx15bgeo/bgeo MEFs were transduced with the indicated factors and selected with G418 for 10 days (white columns) or 16 days (black columns).

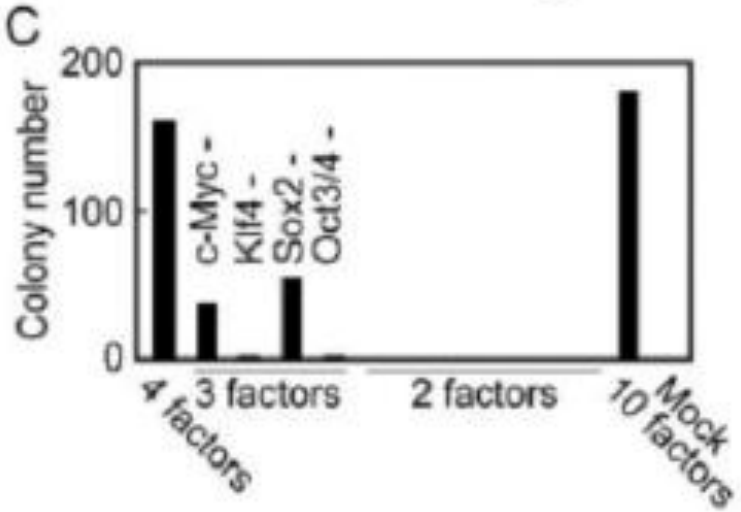
B



Effect of the removal of individual factors from the selected 10 factors on the formation of G418-resistant colonies 16 days after transduction

Effect of the transduction of pools of four, three, and two factors on the formation of G418-resistant colonies 16 days after transduction

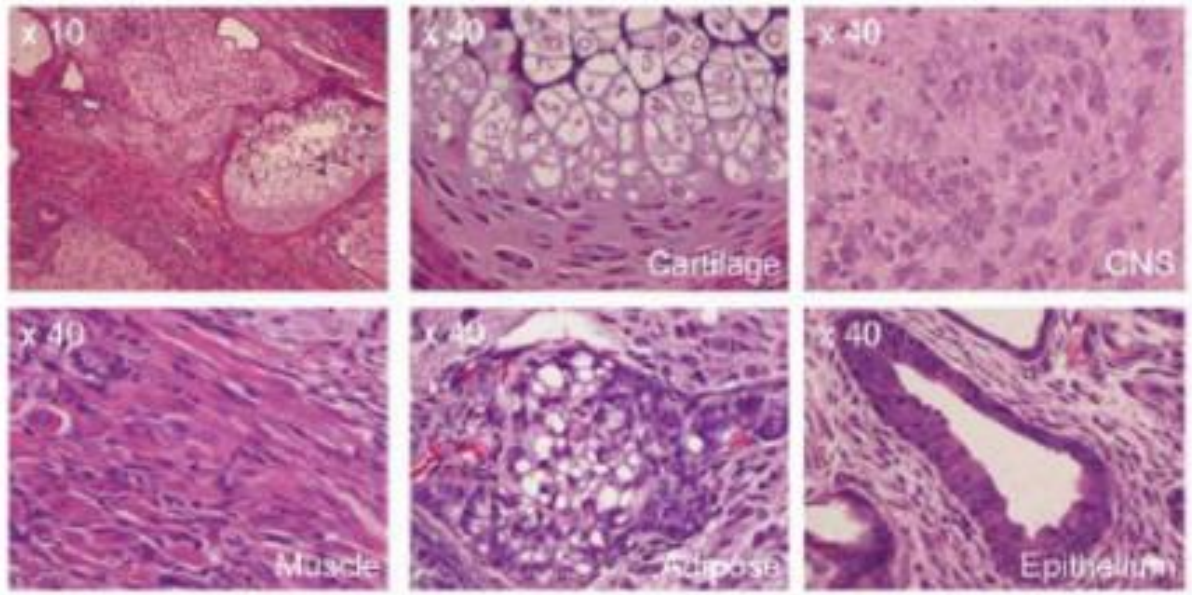
C



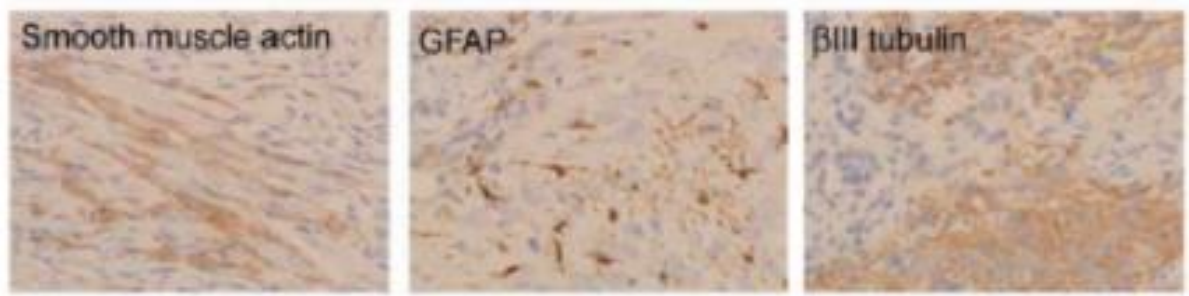
These data demonstrate that iPS cells can be induced from MEF culture by the introduction of four transcription factors, Oct3/4, Sox2, c-Myc, and Klf4.

Pluripotency of iPS Cells Derived from MEFs (mouse embryonic fibroblasts)

(A) Various tissues present in teratomas derived from iPS-MEF4-7 cells.



(B) Immunostaining confirming differentiation into neural tissues and muscles in teratomas derived from iPS-MEF4-7.



Stimulus-triggered fate conversion of somatic cells into pluripotency

Haruko Obokata, Teruhiko Wakayama, Yoshiki Sasai, Koji Kojima, Martin P. Vacanti, Hitoshi Niwa, Masayuki Yamato & Charles A. Vacanti

Affiliations | Contributions | Corresponding authors

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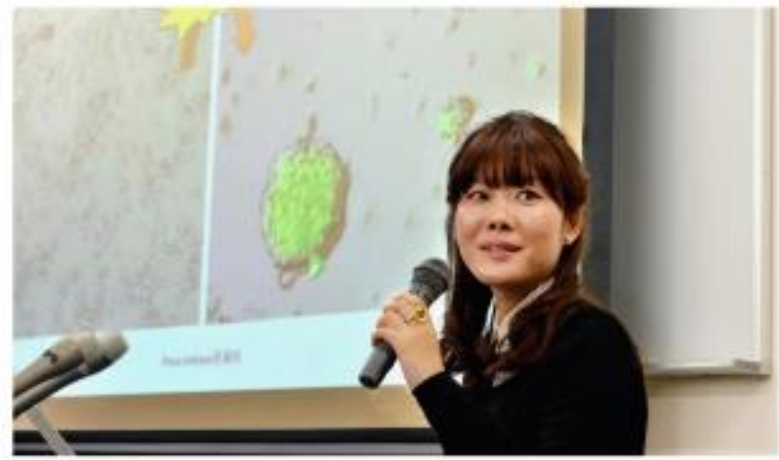


ARTICLE

doi:10.1038/nature12968

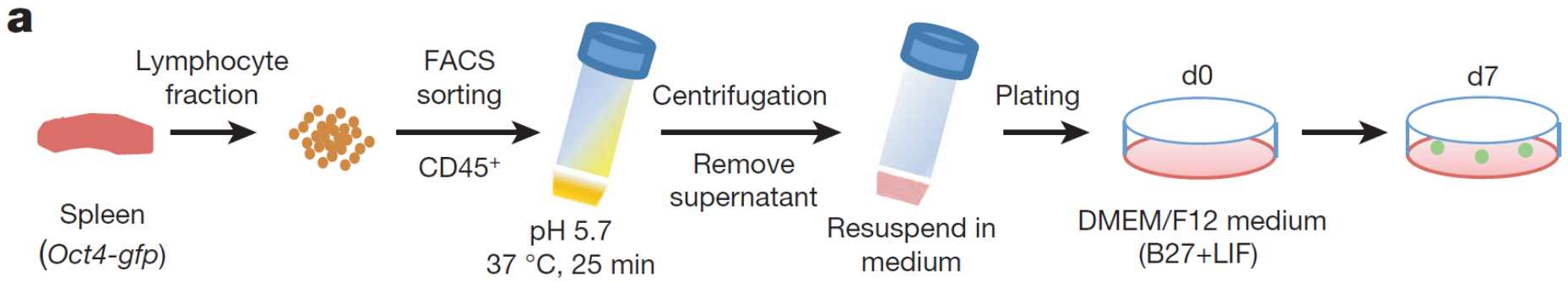
Stimulus-triggered fate conversion of somatic cells into pluripotency

Haruko Obokata[1,2,3], Teruhiko Wakayama[3]†, Yoshiki Sasai[4], Koji Kojima[1], Martin P. Vacanti[1,5], Hitoshi Niwa[6], Masayuki Yamato[7] & Charles A. Vacanti[1]

Here we report a unique cellular reprogramming phenomenon, called stimulus-triggered acquisition of pluripotency (STAP), which requires neither nuclear transfer nor the introduction of transcription factors. In STAP, strong external stimuli such as a transient low-pH stressor reprogrammed mammalian somatic cells, resulting in the generation of pluripotent cells. Through real-time imaging of STAP cells derived from purified lymphocytes, as well as gene rearrangement analysis, we found that committed somatic cells give rise to STAP cells by reprogramming rather than selection. STAP cells showed a substantial decrease in DNA methylation in the regulatory regions of pluripotency marker genes. Blastocyst injection showed that STAP cells efficiently contribute to chimaeric embryos and to offspring via germline transmission. We also demonstrate the derivation of robustly expandable pluripotent cell lines from STAP cells. Thus, our findings indicate that epigenetic fate determination of mammalian cells can be markedly converted in a context-dependent manner by strong environmental cues.

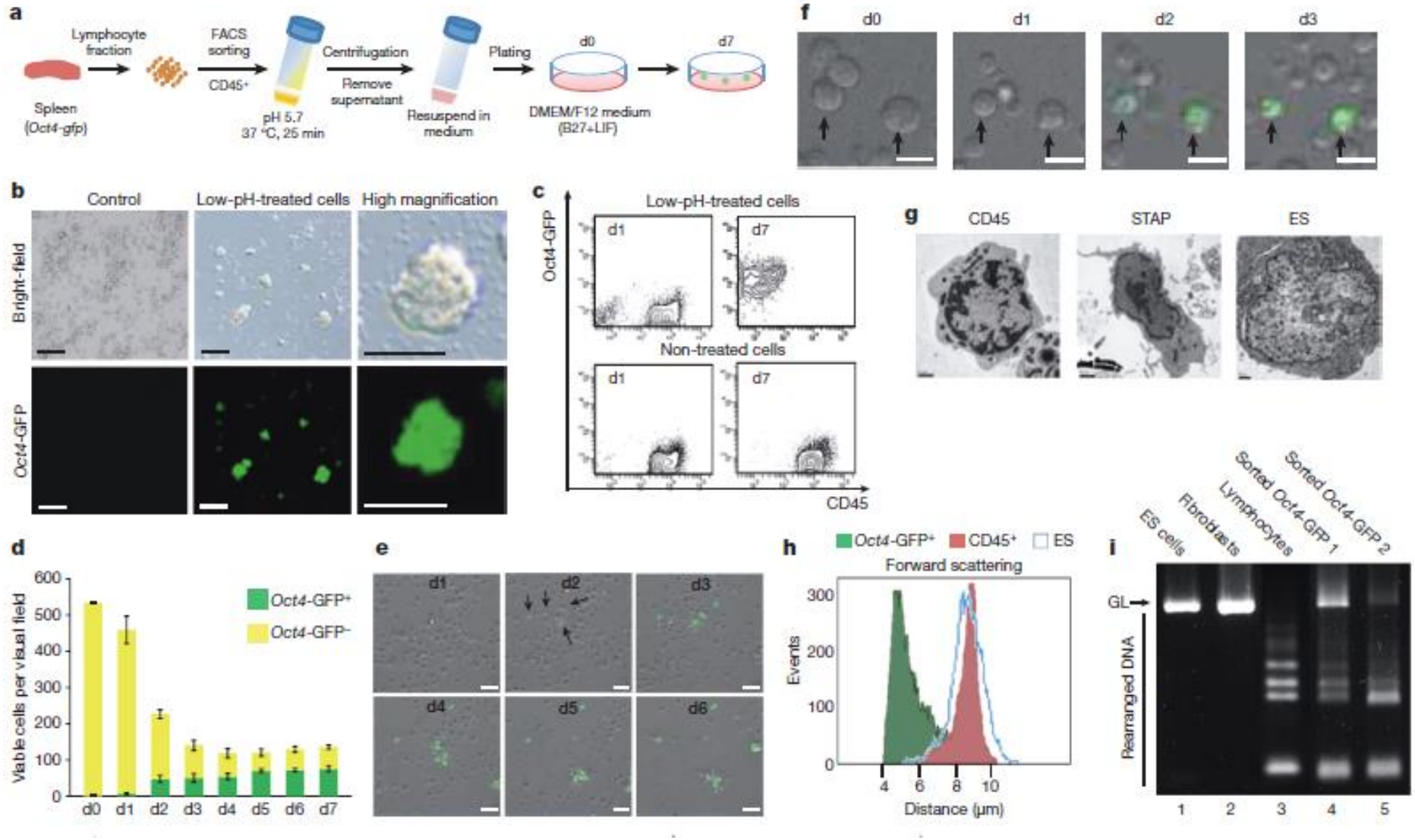
https://www.youtube.com/watch?v=h9QBKborGvI

Stimulus-triggered conversion of lymphocytes into Oct4-GFP+ cells



CD45+ cells were sorted by fluorescence-activated cell sorting (FACS) from the lymphocyte fraction of postnatal spleens (1-week old) of C57BL/6 mice carrying an **Oct4-GFP** transgene, and were exposed to various types of strong, transient, physical and chemical stimuli.
We examined these cells for activation of the Oct4 promoter after culture for several days in suspension using DMEM/F12 medium supplemented with leukaemia inhibitory factor (LIF) and B27.

(LIF is usually added to stem cell culture medium to reduce spontaneous differentiation)



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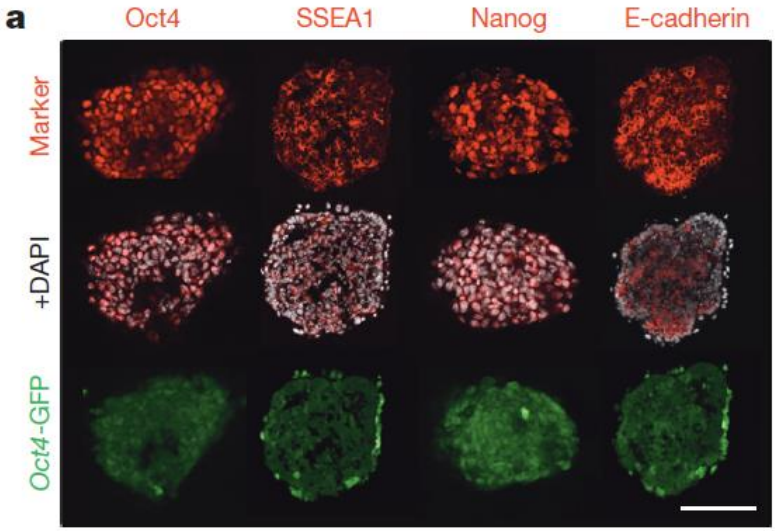
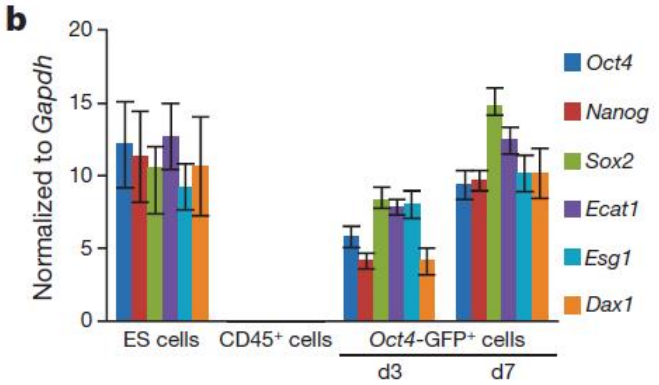
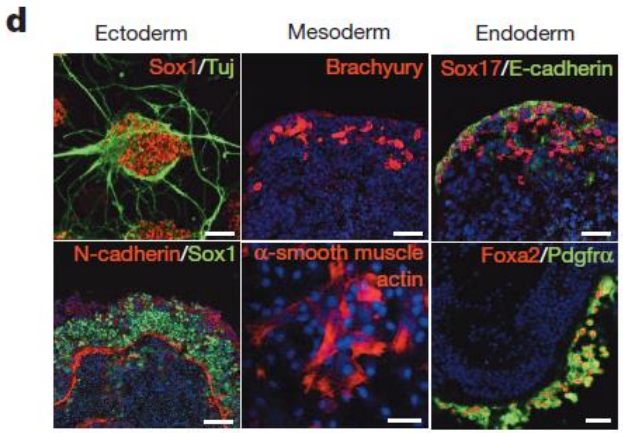
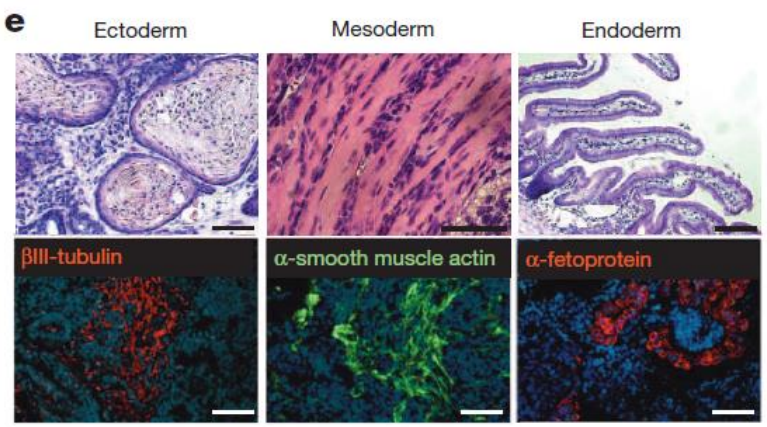




Low-pH-induced Oct4-GFP+ cells represent pluripotent cells.
a, Immunostaining for pluripotent cell markers (red) in day 7 Oct4-GFP1 (green) clusters.
b, qPCR analysis of pluripotency marker genes.
d, Immunostaining analysis of in vitro differentiation capacity of day 7 Oct4-GFP1 cells.
e, Teratoma formation assay of day 7 clusters of Oct4-GFP1 cells.





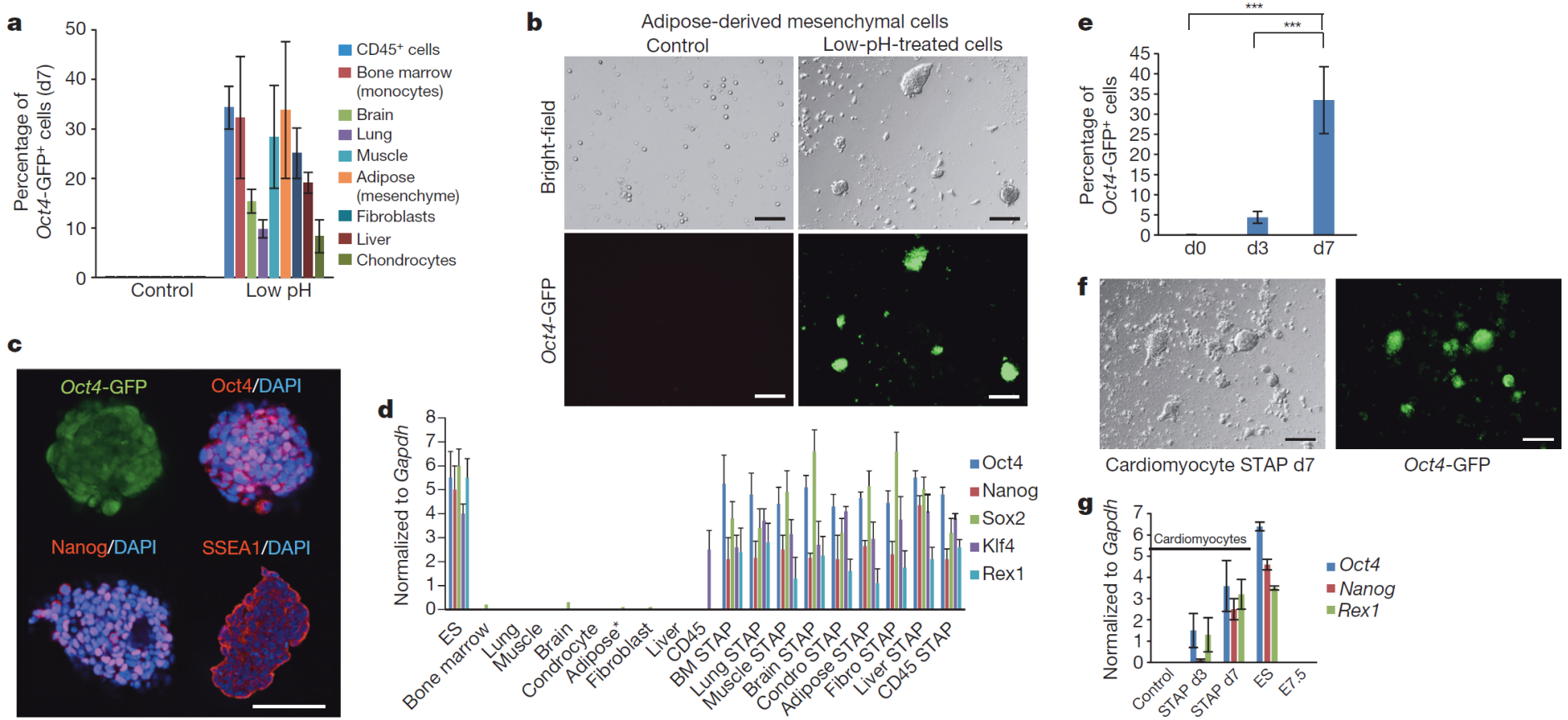
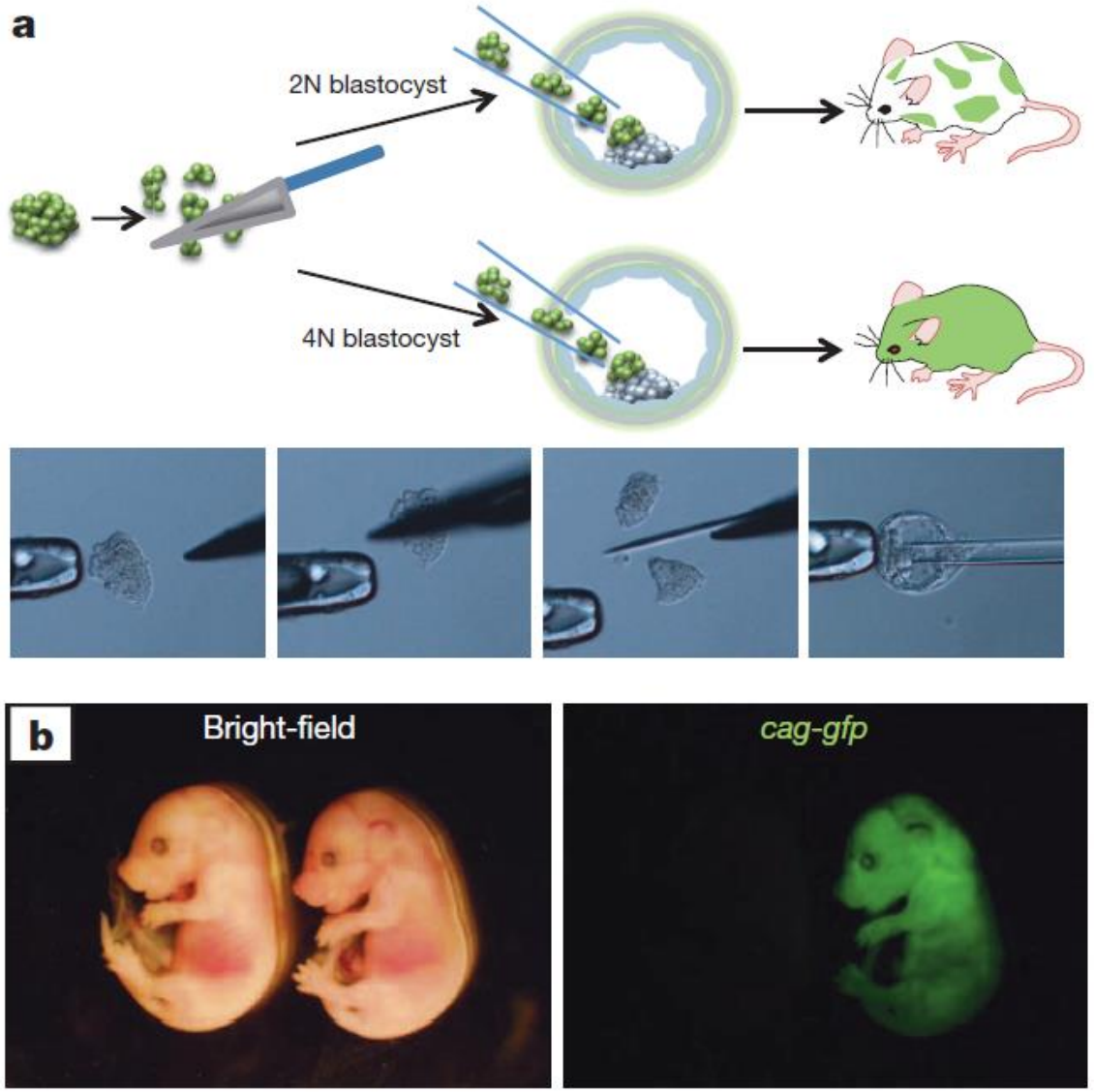


Figure 3 | STAP cell conversion from a variety of cells by low-pH treatment. **a**, Percentage of *Oct4*-GFP$^+$ cells in day 7 culture of low-pH-treated cells from different origins (1×10^5 cells per ml $\times$ 3 ml). The number of surviving cells on day 7 compared to the plating cell number was 20–30%, except for lung, muscle and adipose cells, for which surviving cells were ~10% (n=3, average ± s.d.). **b**, *Oct4*-GFP$^+$ cell clusters were induced by low-pH treatment from adipose-tissue-derived mesenchymal cells on day 7. Scale bar, 100 µm. **c**, Expression of pluripotent cell markers in day 7 clusters of low-pH-treated adipose-tissue-derived mesenchymal cells. Scale bar, 50 µm. **d**, Expression of pluripotency marker genes in STAP cells derived from various tissues. Gene expressions were normalized by *Gapdh* ($n = 3$, average ± s.d.). Asterisk indicates adipose tissue-derived mesenchymal cells. **e**, Quantification of *Oct4*-GFP$^+$ cells in culture of low-pH-treated neonatal cardiac muscle cells. ***$P < 0.001$; Tukey's test ($n = 3$). **f**, Generation of *Oct4*-GFP$^+$ cell clusters (d7) from CD45$^-$ cardiac muscle cells. **g**, qPCR analysis of pluripotency marker genes in STAP cells from CD45$^-$ cardiac muscle cells.



Chimaeric mouse generation from STAP cells.
a, Schematic of chimaeric mouse generation.
b, E13.5 chimaera fetuses from 2N blastocytes injected with STAP cells (derived from B6GFP CD45+ cells carrying **cag-gfp**).

عربي

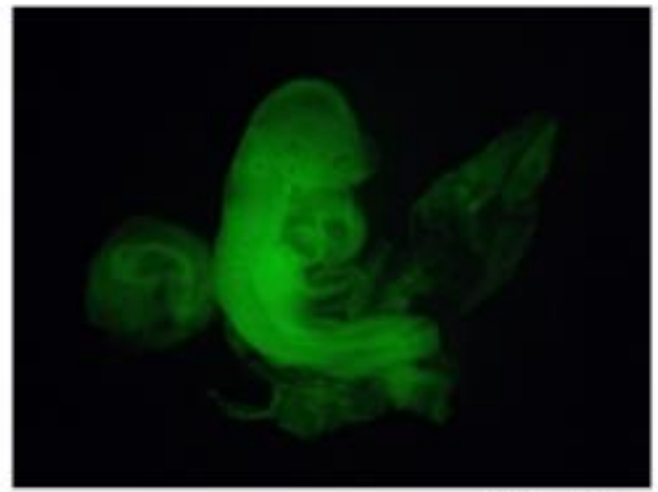
Acid-bath stem-cell study under investigation

Japanese research institute launches inquiry after allegations of irregularities in blockbuster papers.

David Cyranoski

17 February 2014

A leading Japanese research centre has opened an investigation into a groundbreaking stem-cell study after concerns were raised about the work's credibility.



Haruko Obokata

The controversial work involved a mouse embryo injected with cells made pluripotent through stress.

The RIKEN Center for Developmental Biology in Kobe announced on Friday that it is looking into alleged irregularities in the work of biologist Haruko Obokata, who is based at the institution. She shot to fame as the lead author of two papers[1, 2] published in *Nature* last month that demonstrated a way to reprogram mature mouse cells into an embryonic state by simply applying stress, such as exposure to acid conditions or physical pressure on cell membranes. The

Retraction: Stimulus-triggered fate conversion of somatic cells into pluripotency

Haruko Obokata, Teruhiko Wakayama, Yoshiki Sasai, Koji Kojima, Martin P. Vacanti, Hitoshi Niwa, Masayuki Yamato & Charles A. Vacanti

Nature **511**, 112 (03 July 2014) | doi:10.1038/nature13598
Published online 02 July 2014 | Corrected online **23 July 2014**
Article (January, 2014)
Correction (July, 2014)

https://retractionwatch.com/2014/07/02/stap-stem-cell-papers-officially-retracted-as-nature-argues-peer-review-couldnt-have-detected-fatal-problems/

Several critical errors have been found in our Article and Letter (http://dx.doi.org/10.1038/nature12969), which led to an in-depth investigation by the RIKEN Institute. The RIKEN investigation committee has categorized some of the errors as misconduct (see Supplementary Data 1 and Supplementary Data 2). Additional errors identified by the authors that are not discussed in RIKEN's report are listed below.

(1) Figure 1a and b in the Letter both show embryos generated from STAP cells, not a comparison of ES- and STAP-derived chimaeric embryos, as indicated in the legend.

(2) Extended Data Fig. 7d in the Article and Extended Data Fig. 1a in the Letter are different images of the same embryo and not, as indicated in the legends, a diploid chimaera embryo and tetraploid chimaera embryo.

(3) There is an erroneous description in Fig. 1a in the Letter. The right panel of Fig. 1a is not a 'long exposure' image at the camera level but a digitally enhanced one.

(4) In Fig. 4b of the Letter, STAP cell and ES cell are wrongly labelled in a reverse manner.

(5) In the Article, one group of STAP stem cells (STAP-SCs) was reported as being derived from STAP cells induced from spleens of F_1 hybrids from the cross of mouse lines carrying identical *cag-gfp* insertions in chromosome 18 in the background of 129/Sv and B6, respectively, and that they were maintained in the Wakayama laboratory. However, further analysis of the eight STAP-SC lines indicates that, while sharing the same 129×B6 F_1 genetic background, they have a different GFP insertion site. Furthermore, while the mice used for STAP cell induction are homozygous for the GFP transgene, the STAP-SCs are heterozygous. The GFP transgene insertion site matches that of the mice and ES cells kept in the Wakayama laboratory. Thus, there are inexplicable discrepancies in genetic background and transgene insertion sites between the donor mice and the reported STAP-SCs.

We apologize for the mistakes included in the Article and Letter. These multiple errors impair the credibility of the study as a whole and we are unable to say without doubt whether the STAP-SC phenomenon is real. Ongoing studies are investigating this phenomenon afresh, but given the extensive nature of the errors currently found, we consider it appropriate to retract both papers.

STEM CELLS

Bone-marrow-derived cells and heart failure—the debate goes on

Annarosa Leri and Piero Anversa

The therapeutic efficacy of bone-marrow-derived cells in patients with acute or chronic myocardial infarction has been a matter of intense debate. Three new clinical trials—the Swiss-AMI, CELLWAVE, and C-CURE studies—unfortunately do not resolve the controversy in the field of cell therapy for the damaged heart.

Leri, A. & Anversa, P. *Nat. Rev. Cardiol.* **10**, 372–373 (2013); published online 28 May 2013; doi:10.1038/nrcardio.2013.81

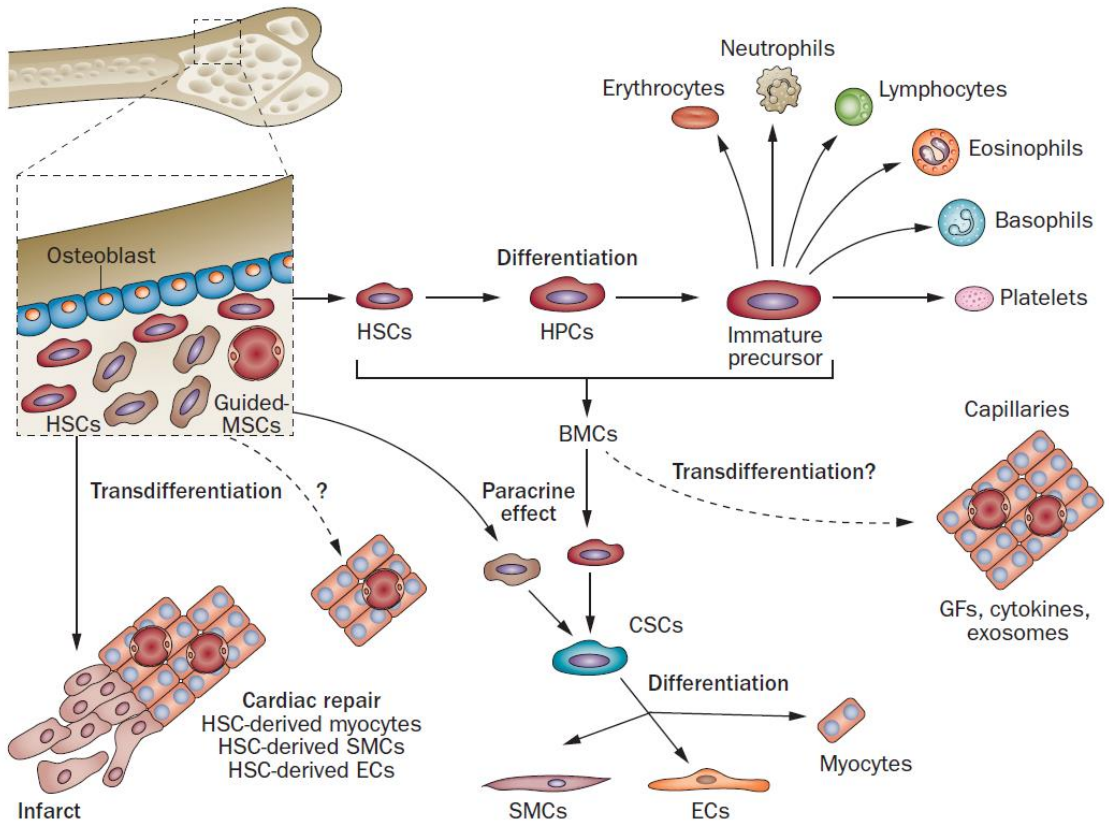


Figure 1 | Several cell types have been implemented clinically to promote myocardial regeneration. The possible mechanisms of action are indicated. Abbreviations: BMC, bone-marrow-derived mononuclear cell; CSC, cardiac stem cell; EC, endothelial cell; GF, growth factor; HPC, haematopoietic progenitor cell; HSC, haematopoietic stem cell; MSC, mesenchymal stromal cell; SMC, smooth muscle cell.

https://retractionwatch.com/2018/12/13/anversa-cardiac-stem-cell-lab-earns-13-retractions/

Harvard Calls for Retraction of Dozens of Studies by Noted Cardiac Researcher

Some 31 studies by Dr. Piero Anversa contain fabricated or falsified data, officials concluded. Dr. Anversa popularized the idea of stem cell treatment for damaged hearts.

https://www.nytimes.com/2018/10/15/health/piero-anversa-fraud-retractions.html

Vol 463|25 February 2010|doi:10.1038/nature08797 nature

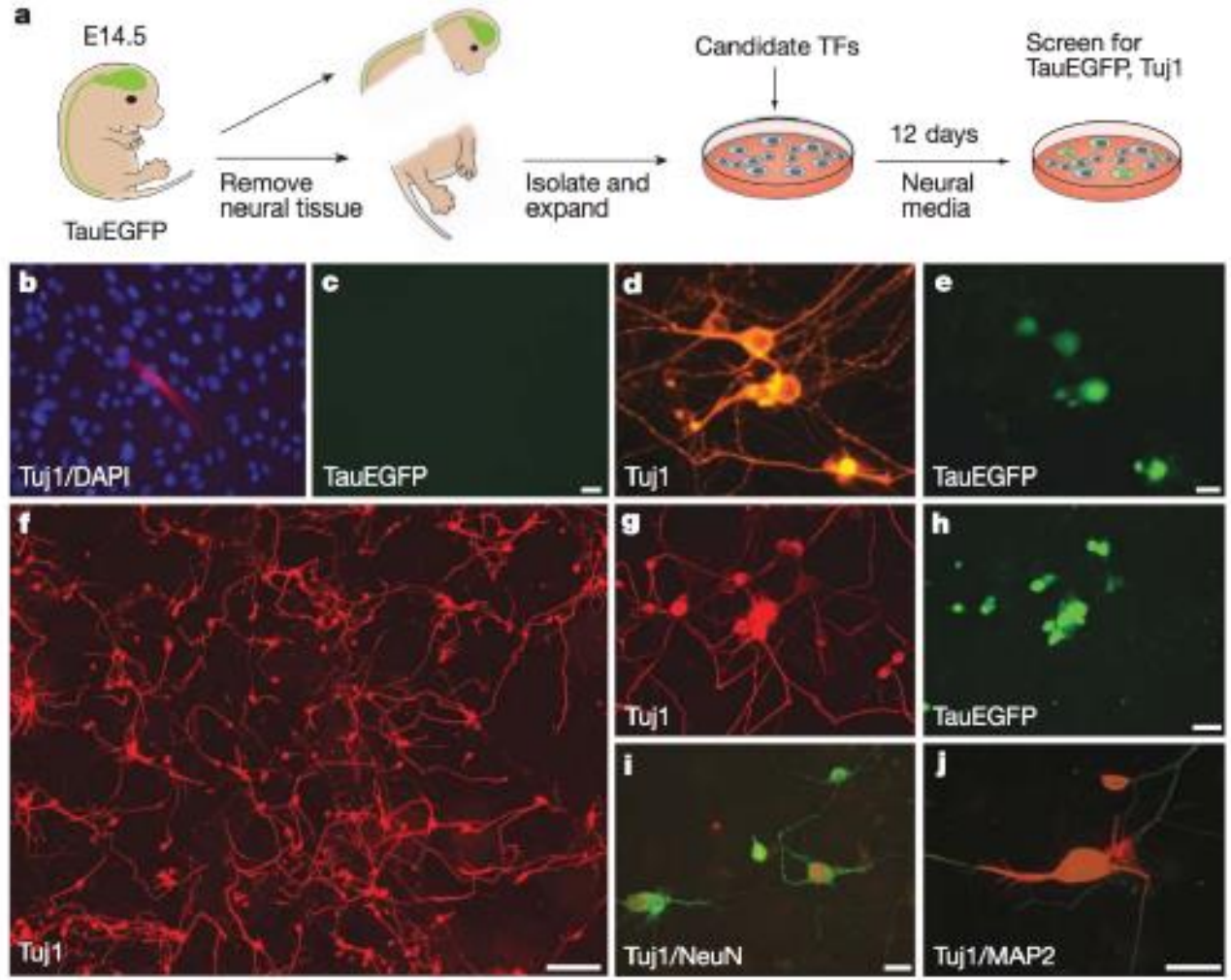
ARTICLES

Direct conversion of fibroblasts to functional neurons by defined factors

Thomas Vierbuchen[1,2], Austin Ostermeier[1,2], Zhiping P. Pang[3], Yuko Kokubu[1], Thomas C. Südhof[3,4] & Marius Wernig[1,2]

Cellular differentiation and lineage commitment are considered to be robust and irreversible processes during development. Recent work has shown that mouse and human fibroblasts can be reprogrammed to a pluripotent state with a combination of four transcription factors. This raised the question of whether transcription factors could directly induce other defined somatic cell fates, and not only an undifferentiated state. We hypothesized that combinatorial expression of neural-lineage-specific transcription factors could directly convert fibroblasts into neurons. Starting from a pool of nineteen candidate genes, we identified a combination of only three factors, *Ascl1*, *Brn2* (also called *Pou3f2*) and *Myt1l*, that suffice to rapidly and efficiently convert mouse embryonic and postnatal fibroblasts into functional neurons *in vitro*. These induced neuronal (iN) cells express multiple neuron-specific proteins, generate action potentials and form functional synapses. Generation of iN cells from non-neural lineages could have important implications for studies of neural development, neurological disease modelling and regenerative medicine.

Starting from a pool of nineteen candidate genes, a combination of only three factors, Ascl1, Brn2 (also called Pou3f2) and Myt1l, that suffice to rapidly and efficiently convert mouse embryonic and postnatal fibroblasts into functional neurons in vitro has been identified.



These induced neuronal (iN) cells express multiple neuron-specific proteins, generate action potentials and form functional synapses.

nature COMMUNICATIONS

ARTICLE

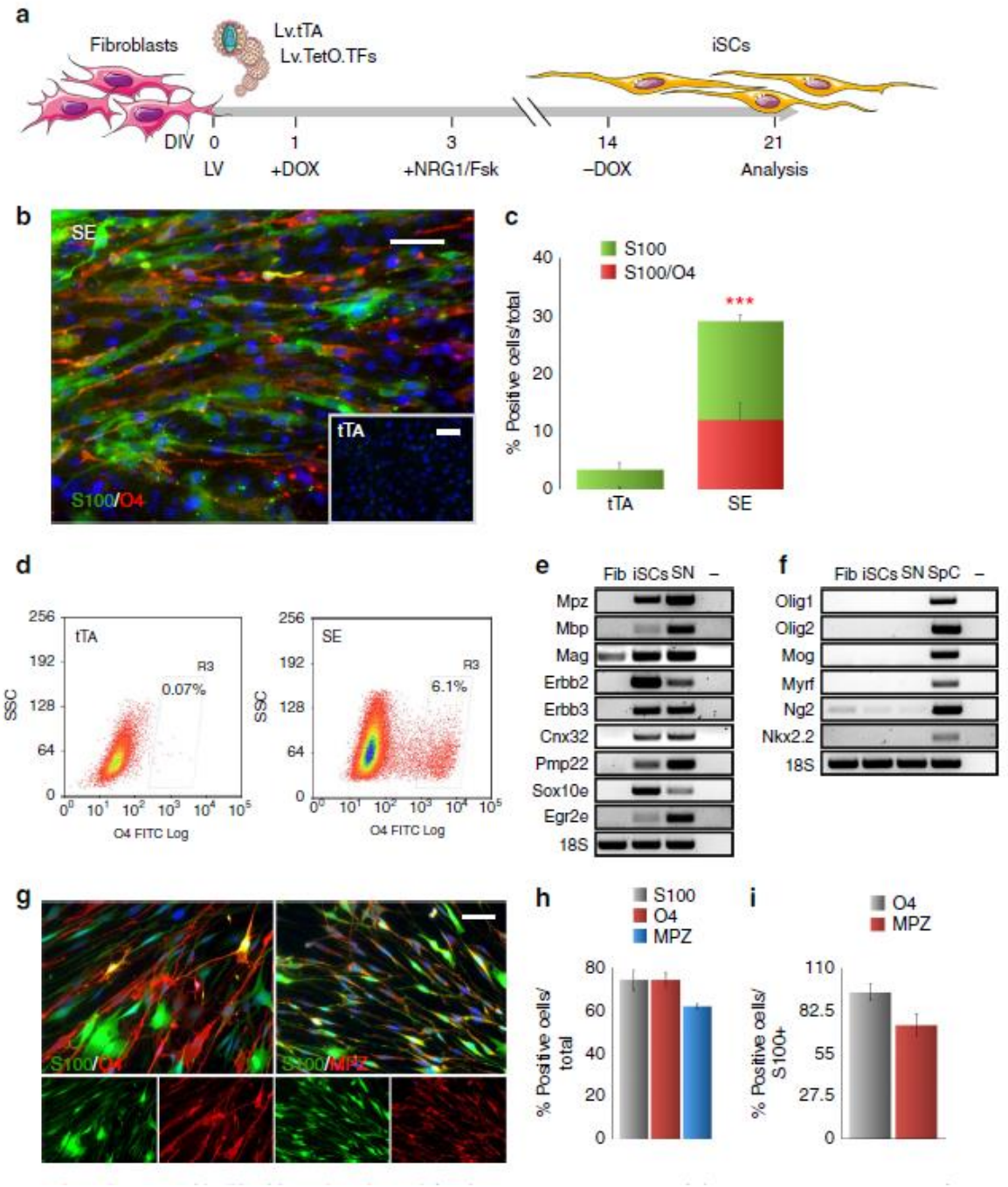
Received 29 Jun 2016 | Accepted 18 Nov 2016 | Published 7 Feb 2017

DOI: 10.1038/ncomms14088 OPEN

Two factor-based reprogramming of rodent and human fibroblasts into Schwann cells

Pietro Giuseppe Mazzara[1,2], Luca Massimino[1], Marta Pellegatta[3], Giulia Ronchi[4], Alessandra Ricca[5], Angelo Iannielli[1], Serena Gea Giannelli[1], Marco Cursi[6], Cinzia Cancellieri[1], Alessandro Sessa[1], Ubaldo Del Carro[6], Angelo Quattrini[7], Stefano Geuna[4], Angela Gritti[5], Carla Taveggia[3] & Vania Broccoli[1,8]

Schwann cells (SCs) generate the myelin wrapping of peripheral nerve axons and are promising candidates for cell therapy. However, to date a renewable source of SCs is lacking. In this study, we show the conversion of skin fibroblasts into induced Schwann cells (iSCs) by driving the expression of two transcription factors, Sox10 and Egr2. iSCs resembled primary SCs in global gene expression profiling and PNS identity. *In vitro*, iSCs wrapped axons generating compact myelin sheaths with regular nodal structures. Conversely, iSCs from Twitcher mice showed a severe loss in their myelinogenic potential, demonstrating that iSCs can be an attractive system for *in vitro* modelling of PNS diseases. The same two factors were sufficient to convert human fibroblasts into iSCs as defined by distinctive molecular and functional traits. Generating iSCs through direct conversion of somatic cells offers opportunities for *in vitro* disease modelling and regenerative therapies.



Direct conversion of mouse skin fibroblasts into iSCs.

(**a**) Schematic representation of the reprogramming strategy for generating iSCs using the dox-inducible lentiviral transgenes expressing tTA, combinations of transcription factors (TFs) together with the small molecules NRG1 and Fsk.

(**b**) S100 (green) and O4 (red) immunofluorescence staining of Sox10/Egr2 (SE) or tTA-infected (inset) MEFs.

(**c**) Quantification of S100β and S100β/O4β cells expressed as percentage over the total number of cells in experimental (SE) and control (tTA) conditions

(**d**) Fluorescence-activated cell sorting-based purification of the O4β iSC population.

(**e**) Expression of SC cardinal markers by RT–PCR in fibroblasts (Fib), induced SCs and adult sciatic nerve (SN, positive control). Sox10e, endogenous Sox10, Egr2e, endogenous Egr2.

f) Expression of Oligodendrocyte-specific markers by RT–PCR in fibroblasts (Fib), induced SCs, sciatic nerve (SN) and P20 spinal cord (SpC, positive control).

(**g**) S100/O4 and S100/MPZ immunofluorescence staining in iSCs.

STEM CELLS

- Embryonic stem cells (ESC)
- Adult stem cells (ASC)

(**physiological** SC that are present at different stages of the life)

- Nuclear transfer stem cells (NTSC)
- Induced pluripotent stem cells (iPS)

(**artificially reprogrammed** SC, by nuclear transfer or “induction”)

- Cancer stem cells (CSC)

(**pathological** SC that are present in cancer)

Adult stem cells

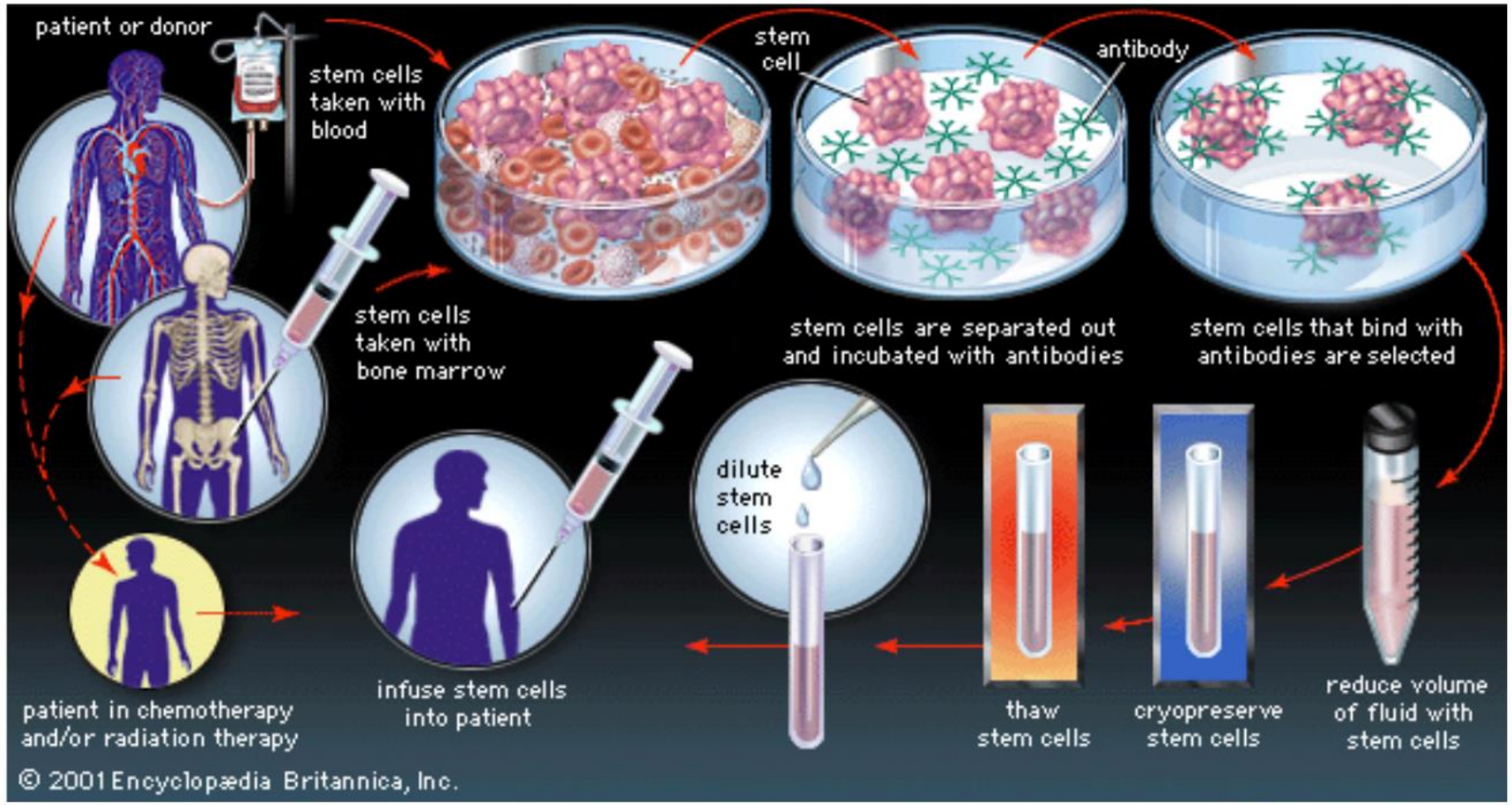
The most extensively studied adult stem cell is the hematopoietic stem cell (HSC). Neural stem cells (NSC) give rise to neurons, astrocytes, and oligodendrocytes. Mesenchymal stem cells (MSC) differentiate into fibroblasts, osteoblasts, chondroblasts, adipocytes, and skeletal muscle. Other stem cells have been identified, including gastrointestinal stem cells, epidermal stem cells, and hepatic stem cells (also called oval cells).

Adult stem cell markers

Hematopoietic stem cells (HSCs)	0·005% of BM CD150+/cKIT+/SCA1+/CD244-neg/CD48-neg/LIN-neg/nestin-neg Less pluripotent progenitors: CD150-neg/CD244+/CD48-neg Committed progenitors: CD150-neg/CD244-neg/CD48+ (Kiel *et al.* 2005)
Mesenchymal stem cells (MSCs)	Nestin+, CD271+/CD146 and CD146+, CD29, CD105 Some markers differ according to tissue of origin Can be located either individually or as the non-parenchymal part of the ASC niche (Zapata 2012)
Muscle stem cells (MuSCs)	Previously known as satellite cells Stemness: PAX3+/PAX7+ and microRNA-489+ (Dek receptor downregulation) Myocyte committed progenitors: MyoD+/Myf5+/Myostatin+ and Dek+ (Rando 2005, Relaix *et al.* 2005, Cheung *et al.* 2012)
Small intestine stem cells	LGR5+high/SOX2+high/SOX9+low BMI+other stem population or different functional state of the same stem population? Committed enteroendocrine precursors: SOX9+high (Barker *et al.* 2007, Sangiorgi & Capecchi 2008, Formeister *et al.* 2009)
Liver and pancreas stem cell	A common stem cell for both organs under discussion Differentiated hepatocytes have capacity for self-renewal Liver stem cell: SOX9+ Pancreas stem cell: exocrine, ductal, and endocrine populations proposed. Also, a common ductal ASC for all tissues has been proposed Other hypotheses do not involve stem cells but self-renewal of committed precursors, e.g. NGN3+-islet-specific precursors (Xu *et al.* 2008, Burke & Tosh 2012)
Testes: progenitor germ cells (PGCs)	RET+/GFRA1+/PLZ+/ETV5+/BCL6B+/NANOG-neg and OCT4low/SOX2very low/SOX9low (Alvarez CV *et al.* unpublished) Sertoli cells: SOX9+-high (Meng *et al.* 2000, Costoya *et al.* 2004, Lee *et al.* 2007, Oatley *et al.* 2007)
Skin stem cells (bulge)	LGR5+/SOX9+/TCF3+ (and others; Blanpain *et al.* 2004, Tumbar *et al.* 2004, Claudinot *et al.* 2005, Fuchs & Horsley 2011)
Neural stem cells (NSCs)	Niches in the subgranular zone of the dentate gyrus of the hippocampus, the subventricular area of the lateral ventricles, the olfactory bulb, and the spinal cord No consensus as to what layer within the niche constitutes the real NSC Radial glia-like cells: SOX2+/MSH1+/BLBP+ are the most probable candidates *in vivo* and *in vitro* (Ellis *et al.* 2004, Merkle *et al.* 2007, Suh *et al.* 2007, Kriegstein & Alvarez-Buylla 2009, Bonaguidi *et al.* 2011)
Lung stem cells (LuSCs)	Many candidates proposed (p63+-basal airways cells, alveolar type II pneumocytes, Clara cells) with repair properties after injury but expressing differentiation markers A new population proposed in humans cKIT+/SOX2+/OCT4+/KLF4+/NANOG+ with *in vivo* and *in vitro* stem properties (Rock *et al.* 2009, Anversa *et al.* 2011, Kajstura *et al.* 2011)
Cardiac stem cells (CaSCs)	Initial descriptions of HSC recruitment to infarction sites started a debate about putative *in situ* CaSC Fused HSC cardiomyocytes have limited capacity for self-renewal Niches of cKIT+ described in human heart with *in vivo* and *in vitro* CaSC properties (Bearzi *et al.* 2007, D'Amario *et al.* 2011)
Pituitary stem cells (GPSs)	GFRA2+/GFRA3+/RET+/PROP+/SOX2+/OCT4+/KLF4+/SOX9+/PROP+/E-Cadherin-high/bCat high Nestin-negative Under discussion which is the most stem cell population: SOX2+/SOX9+ or SOX2+/SOX9− (Fauquier *et al.* 2008, Chen *et al.* 2009, Garcia-Lavandeira *et al.* 2009, 2010, 2012, Castinetti *et al.* 2011)

Hemopoietic stem cell

Bone marrow transplantation



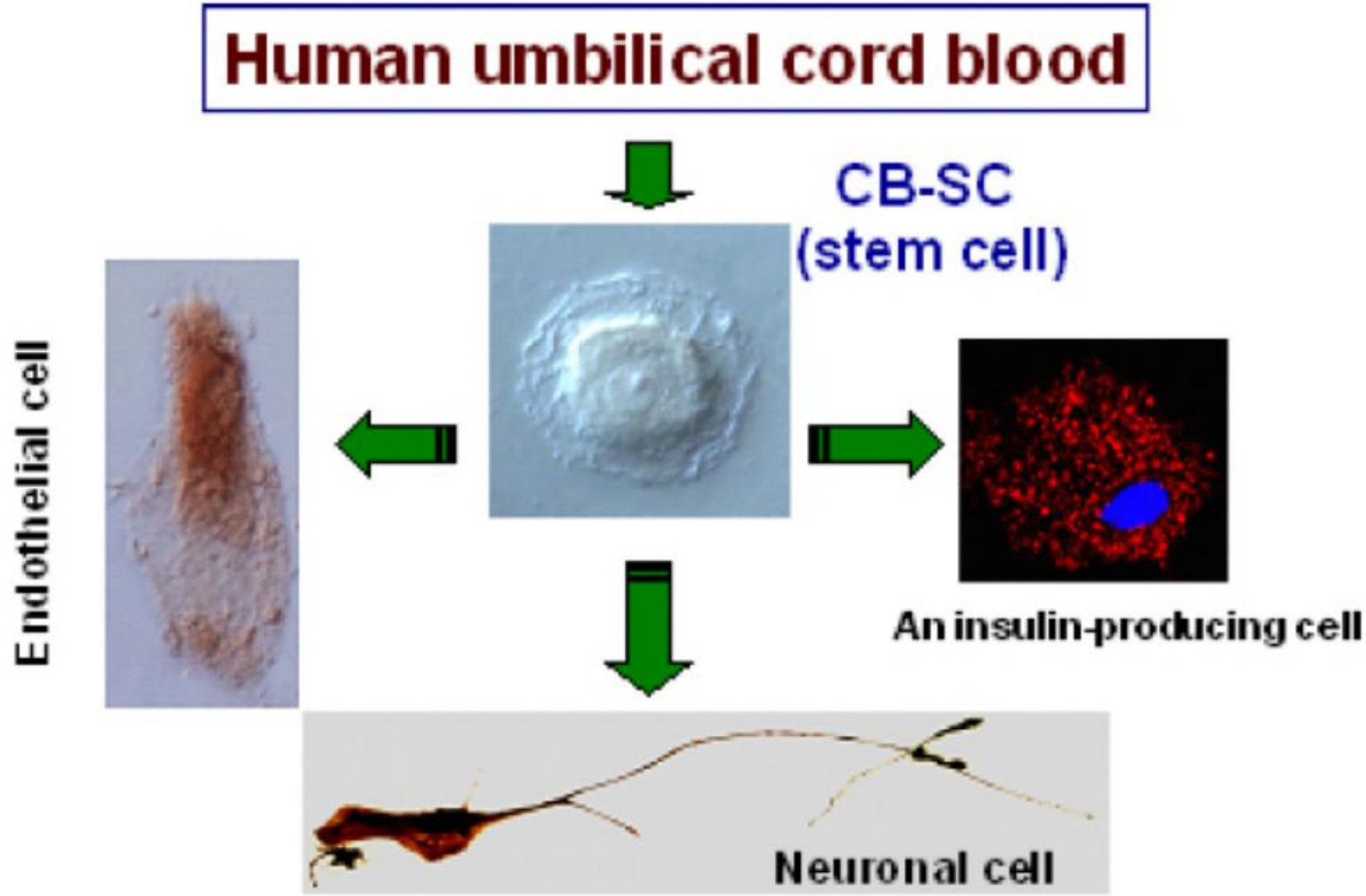


Figure 1. Human umbilical cord blood-derived stem cells (CB-SC) give rise to different cell types. After treatment with different factors, CB-SC turn into insulin-producing cell (right panel: insulin granules showed in red, cell nucleus showed in blue), endothelial cell (left panel), and neuronal cell with long processes (bottom panel), as demonstrated by displaying different specific cell markers and typical cell morphology.