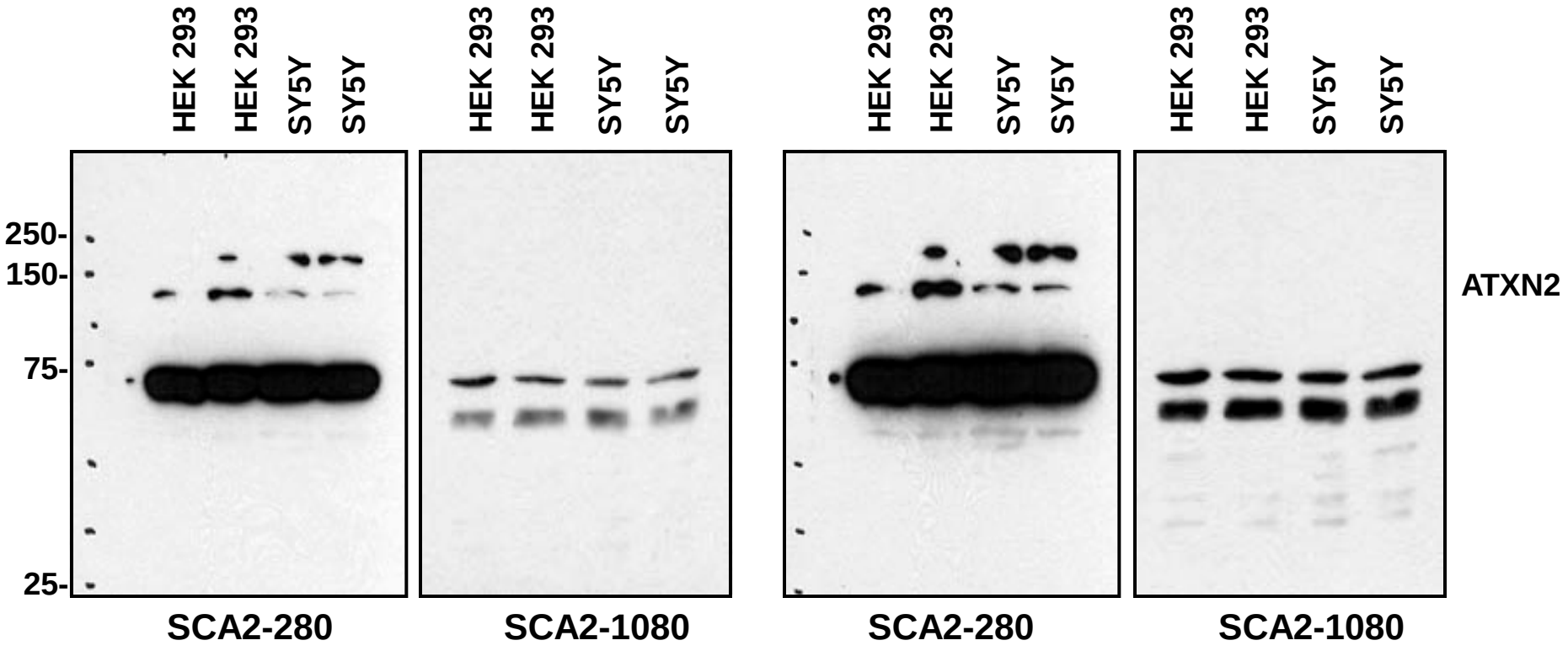
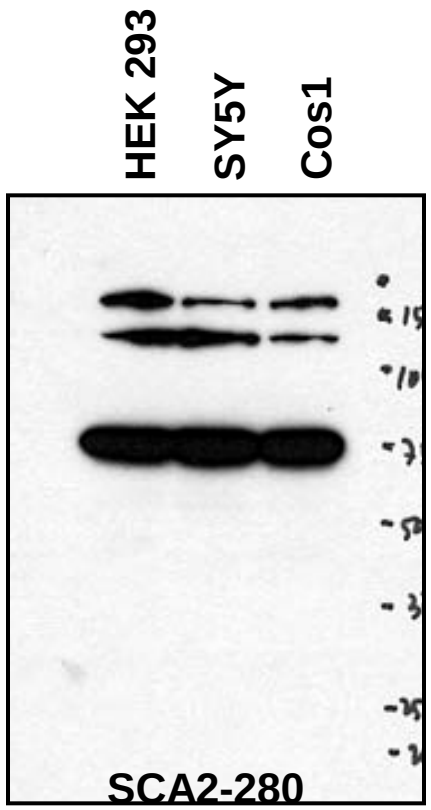
Characterization of SCA2 antibodies

SCA2 (1080) 0.46 ug/ul



Summary of SCA2 Antibodies in Western blot



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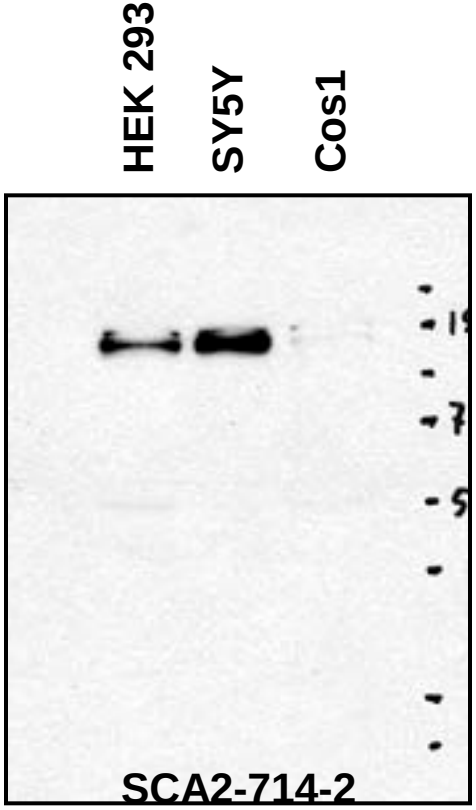

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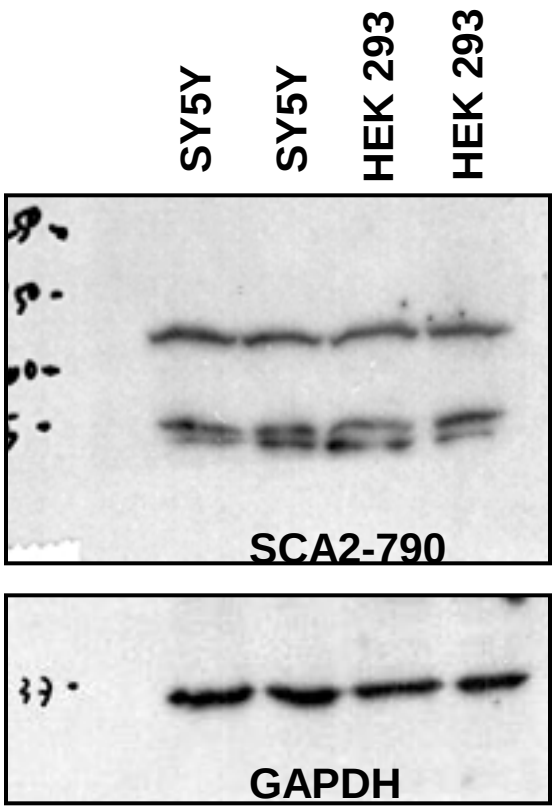


1

Stem Cells

Renew themselves through mitotic cell division

Differentiate into a diverse range of specialized cell types

Types:

Embryonic stem cells (ES)

Pluripotent in nature (Develop into all types of cells in the body)

Adult stem cells

Not pluripotent but they are unipotent or multipotent

Unipotent: produce only one cell type of their own (muscle stem cells).

- **Multipotent: stem cells can differentiate into a number of cells, but only those of a closely related family of cells (hematopoetic stem cells)**

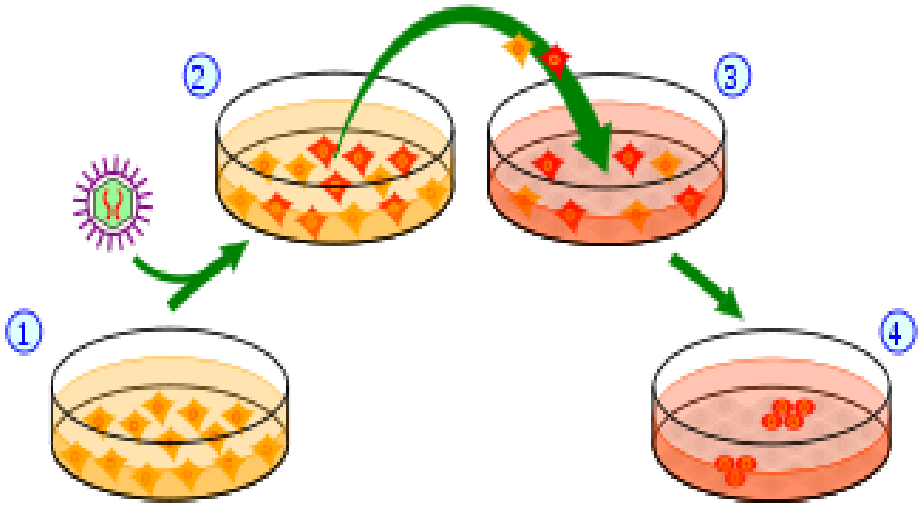
Induced pluripotent stem cells (iPS cells)

Pluripotent stem cells

Derived from a non-pluripoten cell (adult somatic cell) by inducing a "forced" expression of certain genes.

Identical to natural pluripotent stem cells, such as embryonic stem cells

Production of iPSCs



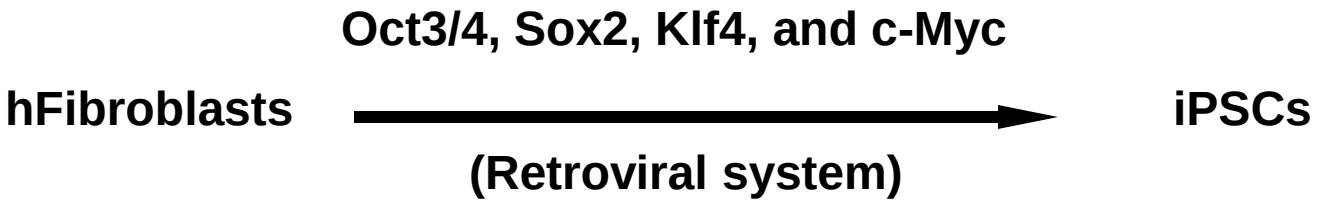
(1) Isolate and culture donor cells.

(2) Transfect stem cell-associated in the genes into the cells by viral vectors.

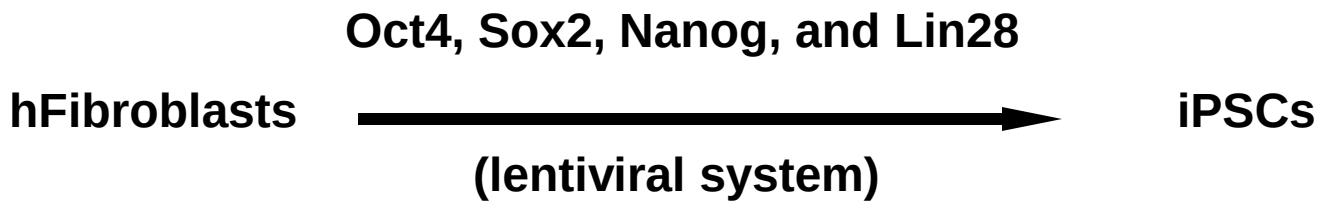
(3) Harvest and culture the cells according to ES cell culture, using mitotically inactivated feeder cells (lightgray).

(4) A small subset of the transfected cells become iPS cells and generate ES-like colonies.

Yamanaka S (Japan)

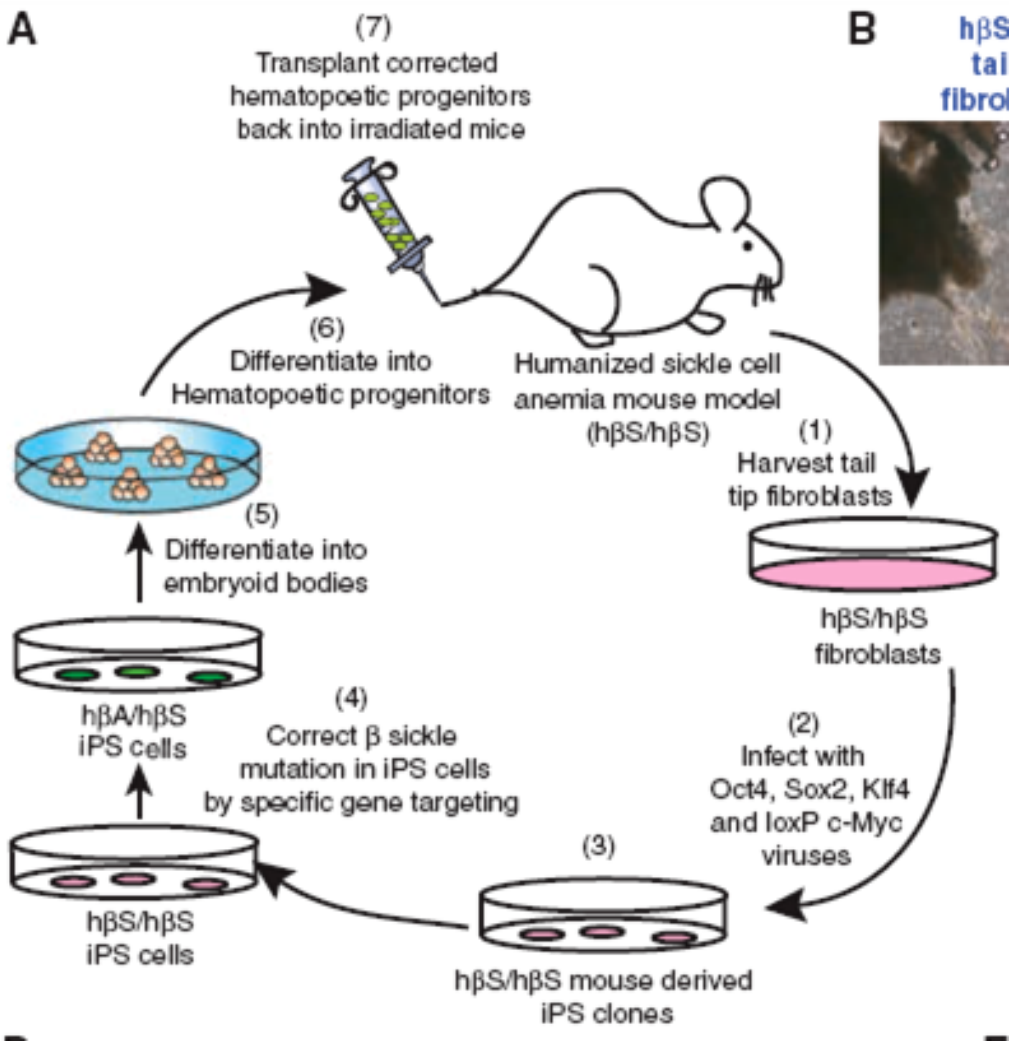


Thomson J (USA)



Treatment of Sickle Cell Anemia Mouse Model with iPS Cells Generated from Autologous Skin

Jacob Hanna, ….Tim M. Townes and Rudolf Jaenisch. Science (318) 1920, 2007



By using a humanized sickle cell anemia mouse model, we show that mice can be rescued after transplantation with hematopoietic progenitors obtained in vitro from autologous iPS cells. Scheme for in vitro reprogramming of skin fibroblasts with defined transcription factors combined with gene and cell therapy to correct sickle cell anemia in mice. Replacement of the hβS gene with a hβA globin gene in sickle iPS cells Homologous recombinantions

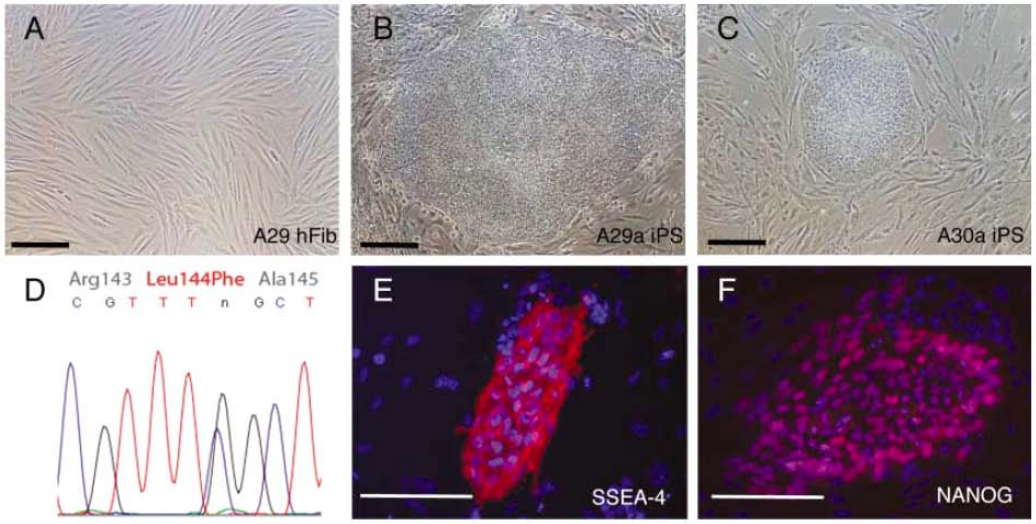
Induced Pluripotent Stem Cells Generated from Patients with ALS Can Be Differentiated into Motor Neurons

John T. Dimos, …. Kevin Eggan. *Science* **321**, 1218 (2008)

ALS is characterized by the progressive degeneration of spinal cord motor neurons

Skin biopsy (ALS patients, 82 and 89 Yrs) → **Fibroblasts** → **Oct4, Sox2, Klf4 and c-Myc** → **iPSCs** (1) → **Differentiation** → **Motor neurons** (2)

(1)

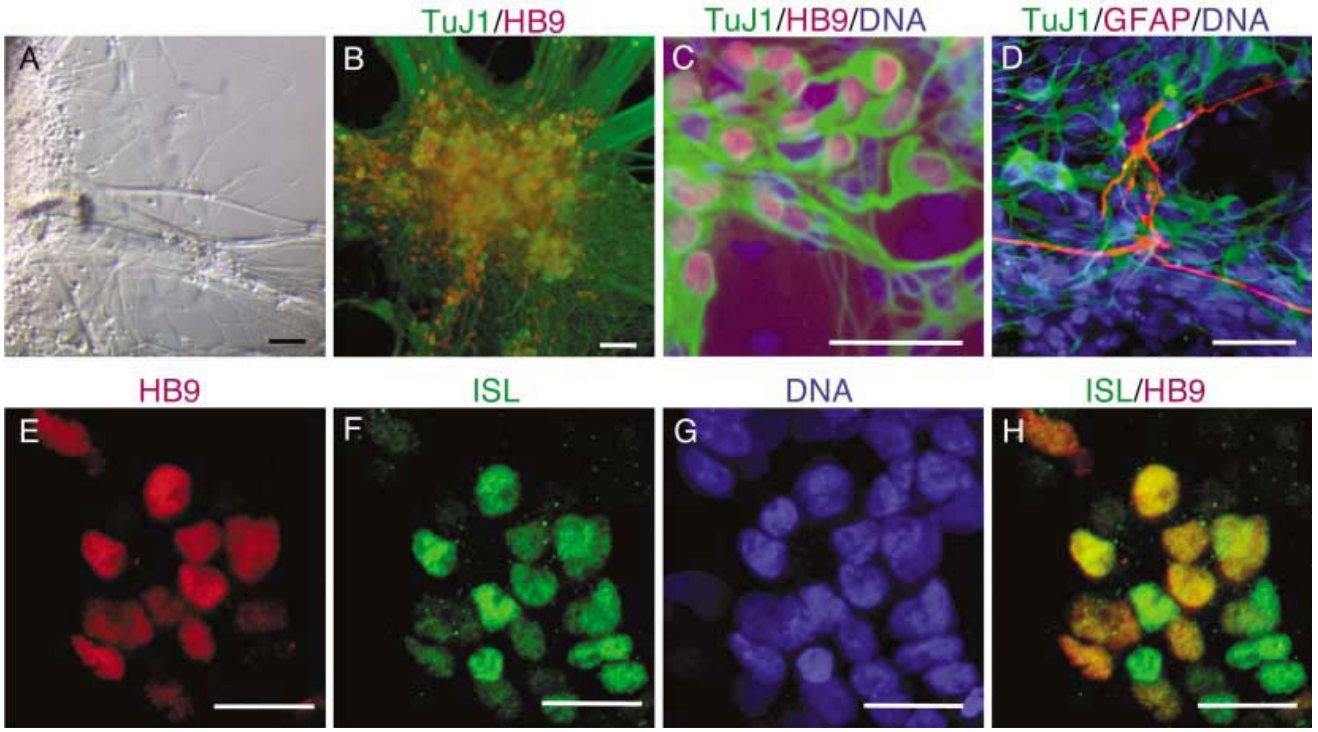


Differentiation

iPSCs → **Motor neurons**

2

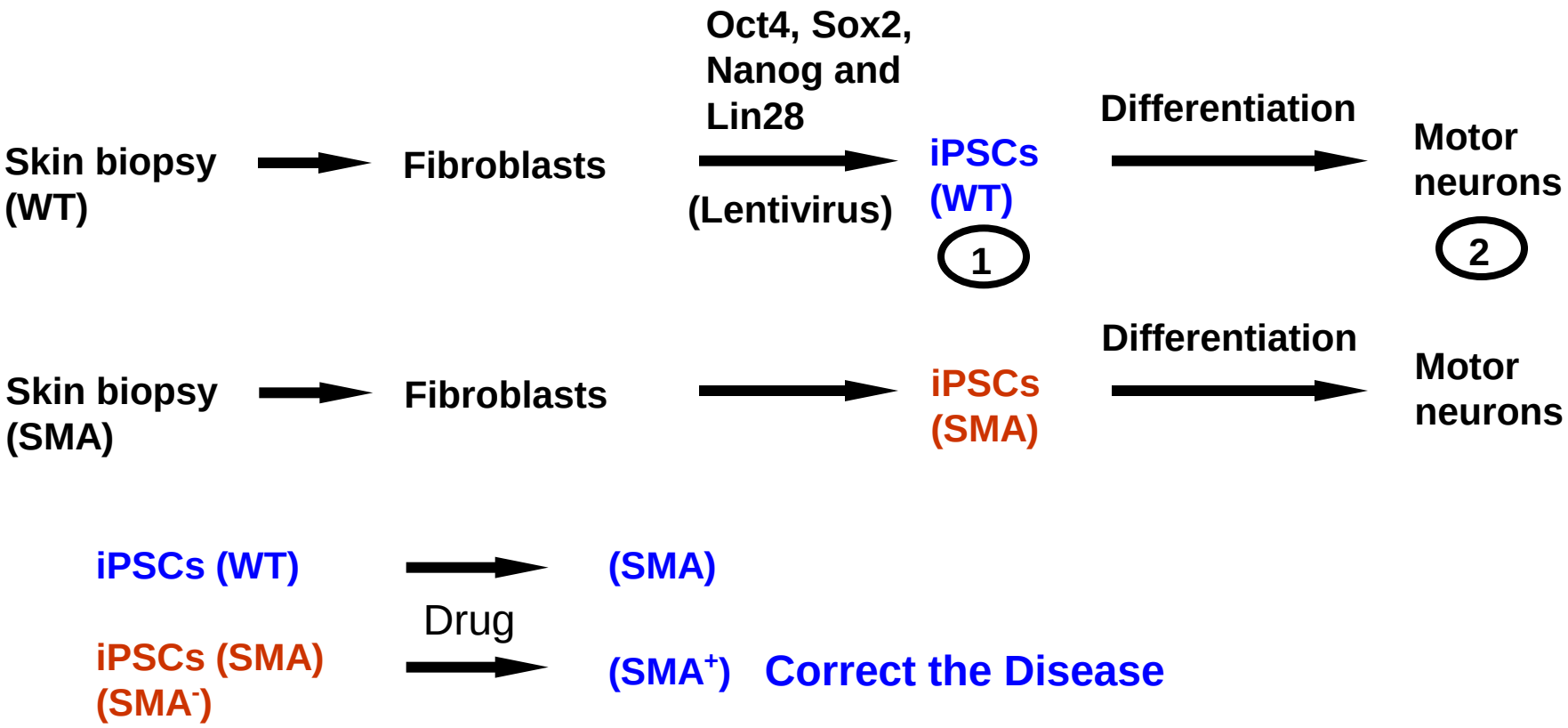
2



Induced pluripotent stem cells from a spinal muscular atrophy patient

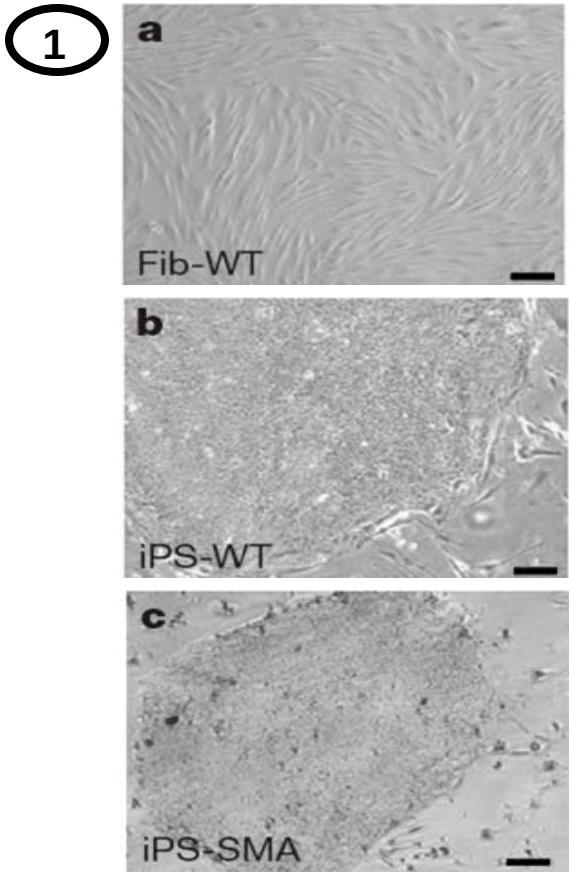
Allison D. Ebert, …Clive N. Svendsen Nature (457) p277; 2009

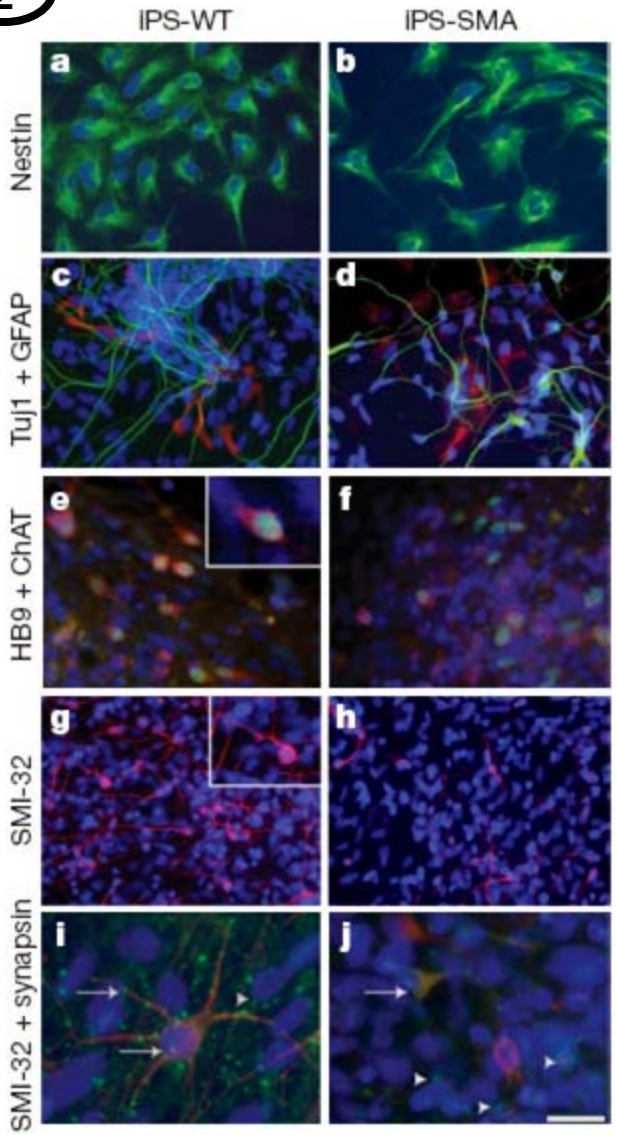
Spinal muscular atrophy (SMA) is an autosomal recessive genetic disorder caused by mutations in the survival motor neuron 1 gene (SMN1) significantly reducing SMN protein expression and resulting in the selective degeneration of lower a-motor neurons.



Skin biopsy → Fibroblasts → (1) iPSCs → Differentiation → Motor neurons

(2)

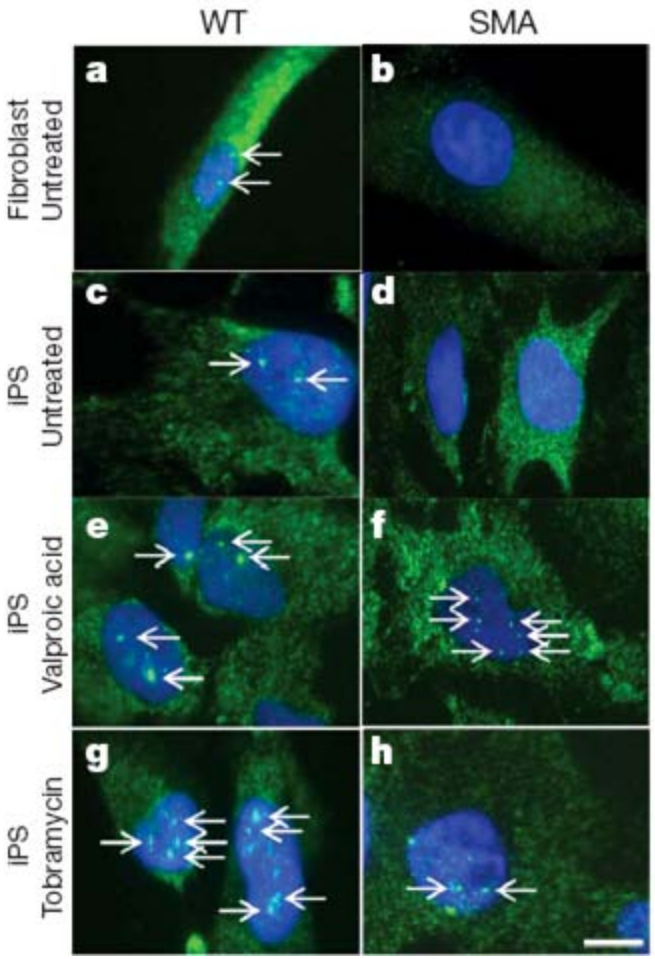


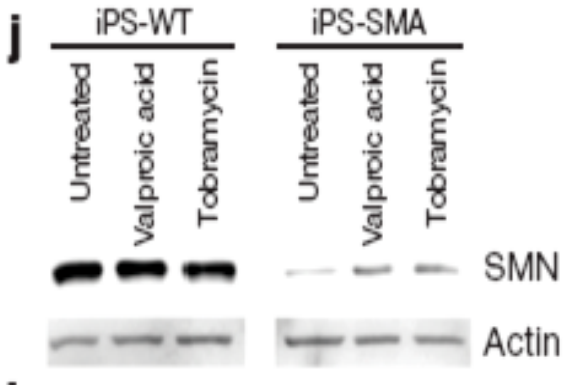


iPSCs (SMA) (SMA^-)

Drug →

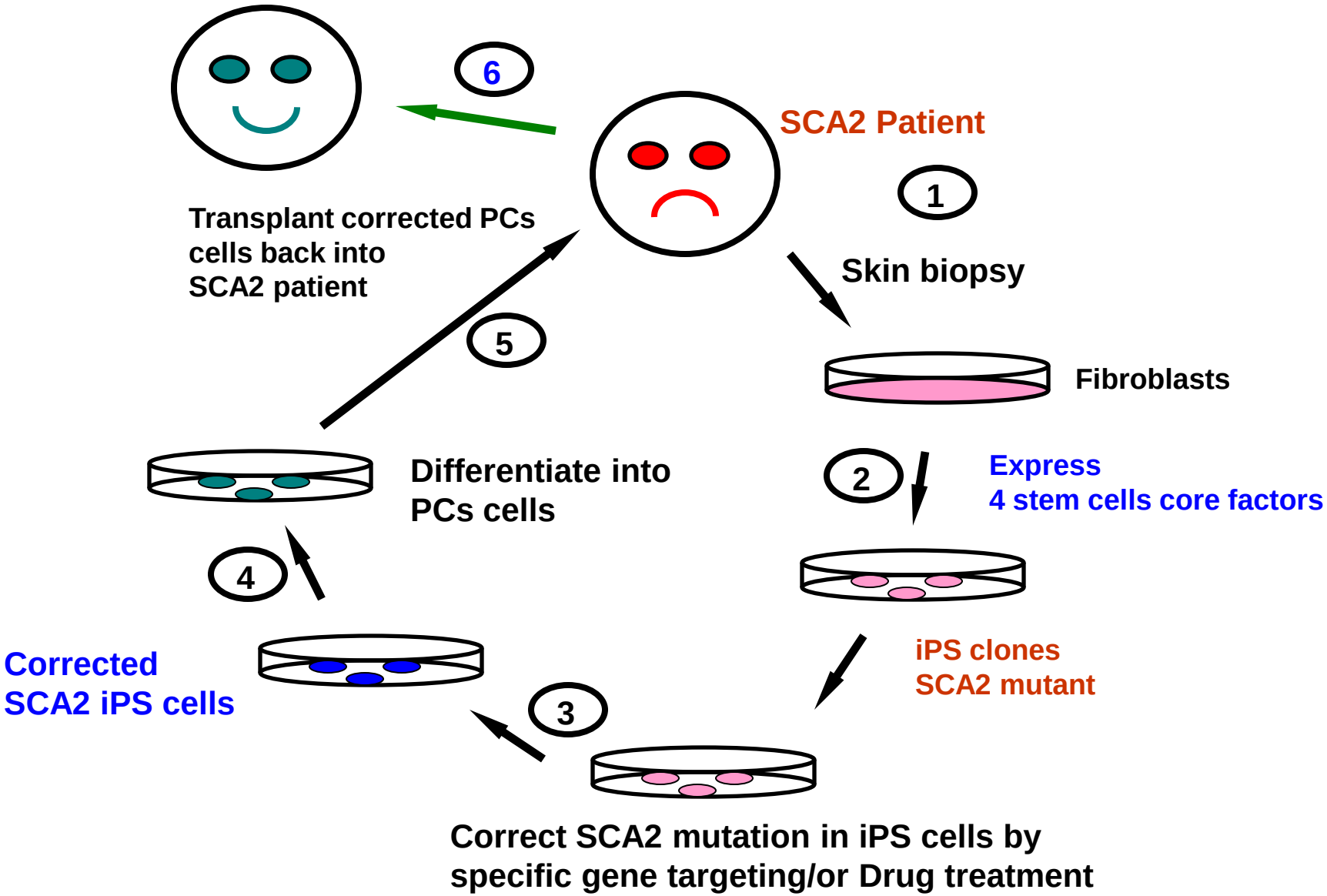
(SMA^+) **Correct the Disease**





iPS-WT and iPS-SMA cells increase SMN protein in response to drug treatment.

Correction of SCA2 patient using iPS cells techniques



Review the Grant proposal

They are trying to establish the cell based therapy to replace the PC in SCA2 patient

Transgenic mice expressing 75Q [(CAG)75] under SCA2 promoter
(Nelso Marino group, Cuba, Neuroscience letter)

Specific aims:

1. **Production of PC from NS (Neural stem) cells.**
2. **Proliferation of PC progenitors and integrate into WT/SCA2 mutant cerebellum**

Ngn2 (neurogenin 2) knock-in inducible mice
At E12.5 stage Ngn2 +ve progenitors give rise to PCs in the adult cerebellum

Fgf8 treatment of proliferating NS cells results in the expression of PC progenitor marker (Ptf1)

Producing/collecting indicator lines for enrichment of commited PC precursors

① **Ngn2-cre-ERT2 mice**

↓ **Tamoxifan**

Ngn2 +ve PC progenitors at E12.5 stage

② **Transgenic mouse; PCP/L7-GFP**

↓

NS cells collection (PC progenitors light up at E15.5 stage

Transplantation

Doner cells

Ngn2- mice/PCP-GFP mice

↓

Cells (VZ) [FACS sorted]

↓

Cells (FACS sorted)

↓ **Transplantation**

WT/SCA2 mutant post natal host (Different ages; P1, P8, P60/adult)

↓ **BrdU injection during 1st week of transplantation**

Sacrifice the mice at different times after BrdU administration (2 hours to 30 days)

↓

Analyze the all parameters